Chapter - 5

Methods for the Analysis of Chromium

OVERVIEW

Chromium is a chemical element in the periodic table that has the symbol Cr and atomic number 24. Chromium is a lustrous, brittle, hard metal. Its colour is silver-gray and it can be highly polished. It is found naturally in rocks, soil, plants and animals, including people. Chromium is present in the environment in several different forms. The most common forms are chromium(0), trivalent (or chromium(III)), and hexavalent (or chromium(VI)). Chromium(III) occurs naturally in the environment and is an essential nutrient required by the human body. In general, chromium(VI) is more toxic than chromium(III).

History

In 1797, Louis Nicolas Vauquelin (French chemist) received samples of crocoite ore (Siberian red lead, PbCrO₄). He was able to produce chromium oxide with a chemical formula of CrO₃, by mixing crocoite with hydrochloric acid. In 1798, Vauquelin discovered that he could isolate metallic chromium by heating the oxide in a charcoal oven. He was also able to detect traces of chromium in precious gemstones, such as ruby, or emerald. During the 1800s chromium was primarily used as a component of paints and in tanning salts. Chromium was named after the Greek word "chroma" meaning colour, because of the many colourful compounds made from it [1].

Notable characteristics

Chromium is a steel-gray, lustrous, hard metal that takes a high polish, and has a high melting point. It is odourless, tasteless and is malleable. Chromium has oxidation states ranging from
chromium(-II) to chromium(+VI). Chromium forms a large number of compounds, in both the chromium(III) and the chromium(VI) forms. Chromium compounds are stable in the trivalent state, with the hexavalent form being the second most stable state. Chromium compounds of oxidation state +6 are powerful oxidants. The chromium(III) compounds are sparingly soluble in water and may be found in water bodies as soluble chromium(III) complexes, while the chromium(VI) compounds are readily soluble in water. Chromium is passivated by oxygen, forming a thin protective oxide surface layer which prevents oxidation of the underlying metal [1,2].

**Occurrence**

Chromium is abundant in nature. It is generally found as trivalent chromium [Cr(III)] or hexavalent chromium [Cr(VI)]. Trivalent chromium occurs naturally in many fresh vegetables, fruits, meat, grains and yeast, and is often added to vitamins as a dietary supplement. Hexavalent chromium is most often produced by industrial processes and may be an indicator of environmental contamination [3]. Chromium is mined as chromite (FeCr$_2$O$_4$) ore. Roughly half the chromite ore in the world is produced in South Africa. Kazakhstan, India and Turkey are also substantial producers. Untapped chromite deposits are plentiful, but geographically concentrated in Kazakhstan and southern Africa. Though native chromium deposits are rare, some native chromium metal has been discovered. The Udachnaya Mine in Russia produces samples of the native metal [1].

**Uses**

Chromium metal is used as an alloying element to harden steel and to manufacture stainless steel. Chromium compounds are used for chrome plating (e.g. protective coatings for automotive and equipment accessories), as dyes, as inorganic paint pigments, for leather tanning, as fungicides and wood preservatives, and as catalysts. More applications are in the photographic industry (sensitiser), in industrial water treatment (including treatment of cooling tower water), as medicinal astringents and antiseptics, and in nuclear and high temperature research. Chromium(III) compounds are employed as pigment, as catalyst, as tanning agent in the tanning industry, in the production of pure chromium metal and chromium(VI) compounds, and in the production of refractory bricks. More specifically, chromium(III) oxide is used as a paint pigment, a fixative for certain textile dyes and as a catalyst. Chromium(III) acetate is used to fix certain textile dyes, to harden photographic emulsions and as a catalyst. Chromium(III) nitrate is used in
the preparation of chrome catalysts, in textile printing operations, and as a corrosion inhibitor. Chromic sulfate is used in tanning, green paints, inks and text dyes and in ceramics [1,2,4].

Toxicity
The toxicity of chromium compounds depends on the oxidation state of the metal. Chromium(VI) is much more toxic than chromium(III), for both acute and chronic exposures. The respiratory tract is the major target organ for chromium(VI) following inhalation exposure in humans. Shortness of breath, coughing and wheezing were reported in cases where an individual inhaled very high concentrations of chromium trioxide. Other effects noted from acute inhalation exposure to very high concentrations of chromium(VI) include gastrointestinal and neurological effects, while dermal exposure causes skin burns in humans. Ingestion of high amounts of chromium(VI) causes gastrointestinal effects in humans and animals, including abdominal pain, vomiting and haemorrhage. Chronic inhalation exposure to chromium(VI) in humans results in effects on the respiratory tract, with perforations and ulcerations of the septum, bronchitis, decreased pulmonary function, pneumonia, asthma, and nasal itching and soreness reported. Chronic human exposure to high levels of chromium(VI) by inhalation or oral exposure may produce effects on the liver, kidney, gastrointestinal and immune systems, and possibly the blood. Chronic exposure to chromium(VI) compounds can cause permanent eye injury, unless properly treated. Dermal exposure to chromium(VI) may cause contact dermatitis, sensitivity, and ulceration of the skin. Exposure to chromium(VI) may result in complications during pregnancy and childbirth. Epidemiological studies of workers have clearly established that inhaled chromium is a human carcinogen, resulting in an increased risk of lung cancer. Although chromium-exposed workers were exposed to both chromium(III) and chromium(VI) compounds, only chromium(VI) has been found to be carcinogenic in animal studies, so United States Environmental Protection Agency (USEPA) has concluded that only chromium(VI) should be classified as a human carcinogen. Animal studies have shown chromium(VI) to cause lung tumors via inhalation exposure. The USEPA has classified chromium(VI) as a Group A, known human carcinogen by the inhalation route of exposure [1,2,5].

Chromium(III) is an essential nutrient required in trace amounts for sugar, protein, and fat metabolism in humans and its deficiency may cause a disease called chromium deficiency. An intake of 50–200 µg of chromium(III) per day is recommended for adults. Acute animal tests have shown chromium(III) to have moderate toxicity from oral exposure [2,5]. People who are allergic to chromium may have asthma attacks after breathing high levels of chromium(III) in air.
Repeated or prolonged skin contact may cause irritation. In severe cases, skin allergy can occur with itching, redness and/or an eczema-like rash [4]. The USEPA has classified chromium(III) as a Group D, not classifiable as to carcinogenicity in humans. The USEPA has stated that "the classification of chromium(VI) as a known human carcinogen raises a concern for the carcinogenic potential of chromium(III)" [5].

**Sources of chromium in the environment**

Chromium is a relatively common element, naturally occurring in rocks, soil, plants, animals, and in volcanic dust and gases. Oil and coal contain traces of chromium(III). Chromium is chiefly found as the chromium(III) form in nature, and rarely as chromium(VI) compounds. Chromium enters the air, water and soil mostly in the chromium(III) and chromium(VI) forms as a result of natural processes and human activities. The most important industrial sources of chromium in the atmosphere are those related to ferrochrome production. Emissions from burning coal and oil, and steel production can increase chromium(III) levels in air. Air emissions of chromium are predominantly of trivalent chromium, and in the form of small particles or aerosols. Stainless steel welding, chemical manufacturing and use of compounds containing chromium(VI) can increase chromium(VI) levels in air. Motor vehicle exhaust may contain chromium. In air, chromium compounds are present mostly as fine dust particles. This dust eventually settles over land and water. Rain and snow help to remove chromium from air. Chromium compounds will usually remain in the air for fewer than 10 days. Although most of the chromium in water binds to dirt and other materials and settles to the bottom, a small amount may dissolve in the water. Most of the chromium in soil does not dissolve easily in water and can attach strongly to the soil. A very small amount of the chromium in soil, however, will dissolve in water and can move deeper in the soil to underground water [2]. Waste streams from electroplating can discharge chromium(VI). Leather tanning and textile industries as well as those that make dyes and pigments can discharge both chromium(III) and chromium(VI) into waterways. Chromium may be oxidised and leached from stainless steel into a water-soluble form [4]. Chromium may exist in water supplies in both the trivalent and hexavalent state, although the trivalent form rarely occurs in potable water [6]. The levels of both chromium(III) and chromium(VI) in soil increase mainly from disposal of commercial products containing chromium, chromium waste from industry and coal ash from electric utilities. Land filling appears to be the most important method for the disposal of chromium wastes generated by chemical industries. Of the total chromium released in the environment by chemical industries, 82.3% is released on land. [2,5].
Human can be exposed to chromium by breathing air, drinking water, or eating food containing chromium or through skin contact with chromium or chromium compounds. The level of chromium in air and water is generally low. Contaminated well or spring water may contain chromium(VI). For the general population, eating foods that contain chromium is the most likely route of chromium(III) exposure. Chromium(III) occurs naturally in many fresh vegetables, fruits, meat, yeast, and grain [2]. Various methods of processing, storage and preparation can alter the chromium content of food. Acidic foods in contact with stainless steel cans or cooking utensils might contain higher levels of chromium because of leaching from stainless steel.

The demand for trace analysis of chromium is particularly high in environmental sciences. Owing to this, many methods for the determination of chromium at trace levels have been developed. Various instrumental methods, such as spectrophotometry, chemiluminescence, AAS, ICP-AES, Mass spectrometry, NAA, X-ray fluorescence etc. have been discussed by Lobinski and Marczenko [7] for the determination of chromium in environmental samples. Spectrophotometric methods still occupied a key role due to their simplicity, reliability, availability of a number of chromogenic reagents, rapidity and determination in the presence of large quantity of associated ions. In this chapter, simple and cost-effective spectrophotometric methods for the determination of chromium and their applications have been described.

**LITERATURE LOOK AT**

Many authors have reported a wide variety of reagents for the spectrophotometric determination of chromium. Chromium is usually determined by the sensitive and fairly selective diphenylcarbazide (DPC) method ($\varepsilon = 4.3 \times 10^4$ L mol$^{-1}$ cm$^{-1}$ at 545 nm) [6,8]. This method, which is more than one hundred times more sensitive than those based on the colours of the Cr(VI) ions, is particularly useful for determining traces of chromium. It is highly susceptible to Fe(III), V(V), Mo(VI), Cu(II) and Hg(II). The DPC has been utilized by many investigators for the spectrophotometric determination of chromium in various samples of matrices [9–14]. Several chromogenic reagents proposed for chromium determination till 1986, have been reviewed by Marczenko [8]. Ressalan et al. [15] have reviewed the utility of chromogenic reagents for the spectrophotometric determination of chromium, reported between 1967 and 1996. Gomez and Callao [16] have described the requirements for chromium determination and speciation and reviewed the analytical methods that have been used in these studies focusing in particular on developments since 2000. Recently reported various spectrophotometric methods for the determination of chromium have been reviewed in the following paragraphs.
A number of azo reagents have been found application for the determination of chromium. Rathiaiah et al. [17] have proposed 2-(5-bromo-2-pyridylazo)-5-diethylaminophenol [5-Br-PADAP] in the presence of benzoate and sodium dodecyl sulphate for the spectrophotometric determination of Cr(III). The red complex has a molar absorption coefficient of \(7.93 \times 10^4\) L mol\(^{-1}\) cm\(^{-1}\) at 595 nm. The same reagent was used by Zhao et al. [18] for the determination of Cr(VI) in an acid medium. Zhao and Han [19] have described a procedure for the spectrophotometric determination of Cr(III) and Cr(VI), when present together by using 5-Br-PADAP. The reaction was accelerated by sodium dodecyl sulfate (SDS) and sodium benzoate (pH = 5 - 5.8) caused a further increase in the absorbance of the chelate. The chelate exhibits maximum absorbance at 590 nm, obeys Beer's law over the concentration range 0.02-0.56 μg/mL of Cr(III), has molar absorption coefficient of \(7.8 \times 10^4\) L mol\(^{-1}\) cm\(^{-1}\). The procedure required heating for colour development. The method was applied to the analysis of Cr(III) in tap water. Anton et al. [20] have studied the complexation reaction of Cr(III) with 2-(5-chloro-2-pyridylazo)-5-dimethylaminophenol (5-Cl-PADAP). The reaction is sensitive (\(\varepsilon = 1.7 \times 10^5\) L mol\(^{-1}\) cm\(^{-1}\)) but, it is pH dependent and requires heating. A spectrophotometric determination of Cr(III), in which Cr(III) forms complex with n-sulfocholorophosphonazo was developed by Zou et al.[21]. The formed complex has molar ratio of 1:1 in HAc-NaAc buffer solution (pH 4.6) and the complex showed a molar absorption coefficient of \(2.99 \times 10^4\) L mol\(^{-1}\) cm\(^{-1}\) at 622 nm. The method was applied to the determination of Cr in steel and human hair samples. De Carvalho et al. [22] have determined Cr(III) with 4-(2-thiazolyazo)-resorcinol (TAR). Cr(III) slowly forms a red complex with TAR at pH 5.7. The complex formation was accelerated by irradiating the reaction mixture with microwave energy. N-cetyl-N, N, N-trimethylammonium bromide (CTAB) was used for increasing the sensitivity of the system (\(\varepsilon = 2.72 \times 10^4\) L mol\(^{-1}\) cm\(^{-1}\) at 545 nm). The method suffers from Fe(III) interference. In another method [23], sulphonazo-chromotropic acid reagent was utilized for the determination of chromium.

Recently, Zaitoun [24] has reported a spectrophotometric determination of chromium(VI) based on the absorbance of its complex with 1,4,8,11-tetraazacyclotetradecane (cyclam). The complex showed a molar absorptivity of \(1.5 \times 10^4\) L mol\(^{-1}\) cm\(^{-1}\) at 379 nm. It is pH dependent and requires a reaction time of 10 min. Determination of chromium by bleaching arsenazo M in H\(_2\)SO\(_4\) medium was described by Huang and Yu-guang [25]. The molar absorption coefficient was found to be \(2.33 \times 10^4\) L mol\(^{-1}\) cm\(^{-1}\) at 550 nm. The method has been applied to the determination of trace chromium in soil.
Jarosz and Biernat [26] have developed a spectrophotometric method for the determination of chromium with Eriochrome Cyanine R in presence of benzylhexadecyl- dimethylammonium chloride as a surfactant. The absorbance of the red-coloured complex was measured at 520 nm ($\varepsilon = 6.8 \times 10^4$ L mol$^{-1}$ cm$^{-1}$). Manganese, silver, chloride and nitrite ions interfered. Liu et al. [27] have used $m$-sulphoantipyrylazo (m-SAA) in the presence of Triton X-100 for the determination of Cr(III). In this method Cr(III) reacts with m-SAA to form a stable 1:1 complex ($\varepsilon = 4.48 \times 10^4$ L mol$^{-1}$ cm$^{-1}$ at 624 nm) in HAc-NaAc medium (pH = 4.0). The reaction required heating. This method was applied to the determination of Cr(III) in synthetic H$_2$O and environmental water samples. A method [28] was proposed based on the formation of ternary complex of chromium(VI) with $o$-chlorophenylfluorone and tetradecylpyridinium chloride, which can be collected on a nitrocellulose membrane. The membrane is dissolved in dimethyl sulphoxide acidified with sulphuric acid, and the absorbance of the resulting complex was measured at 545 nm ($\varepsilon = 1.2 \times 10^5$ L mol$^{-1}$ cm$^{-1}$). The method was applied to the analysis of chromium in different water samples spectrophotometrically.

Methods based on the oxidation of reagents by Cr(VI) have been recommended. Zhao et al. [29] proposed chromotropic acid ($\varepsilon = 1.76 \times 10^4$ L mol$^{-1}$ cm$^{-1}$ at 355 nm). Revanasiddappa and Kiran Kumar [30] have described the oxidation of leuco Xylene Cyanol FF (LXCFF) to its blue form of Xylene Cyanol FF by Cr(VI) in H$_2$SO$_4$ medium. The absorbance of the formed dye was measured in an acetate buffer medium (pH 3.0-4.6) at 615 nm ($\varepsilon = 8.23 \times 10^4$ L mol$^{-1}$ cm$^{-1}$). This method requires heating (∼ 90°C) for 15 min. Recently, a spectrophotometric determination of chromium(VI) with Magneson I [31] has been reported. The method involves the oxidation of Magneson I by Cr(VI), there by a decrease in absorbance of the coloured Magneson I was measured at 420 nm ($\varepsilon = 1.02 \times 10^5$ L mol$^{-1}$ cm$^{-1}$).

Many phenothiazine derivatives such as, thioridazine dihydrochloride ($\varepsilon = 2.577 \times 10^4$ L mol$^{-1}$ cm$^{-1}$ at 640 nm) [32], methdilazine hydrochloride [33], perphenazine ($\varepsilon = 1.87 \times 10^4$ L mol$^{-1}$ cm$^{-1}$ at 526 nm) [34], chlorpromazine hydrochloride ($\varepsilon = 3.28 \times 10^4$ L mol$^{-1}$ cm$^{-1}$) [35] and trifluoperazine hydrochloride [36] have been proposed for the determination of chromium. The above methods are more susceptible to the other redox species, and have been employed to the determination of chromium in soil, ores, steels and other matrices.

Some organic reagents have been utilized for the spectrophotometric determination of chromium, viz, acid chrome blue K [37], chlorophosphonazo I ($\varepsilon = 1.33 \times 10^4$ L mol$^{-1}$ cm$^{-1}$ at 580 nm) [38], dibromoalizarin violet ($\varepsilon = 4.5 \times 10^4$ L mol$^{-1}$ cm$^{-1}$ at 620 nm) [39], flavonoid dyes [40], 4,5-dibromophenylfluorone and polysorbate 60 [41], malachite green ($\varepsilon = 8.0 \times 10^4$
L mol$^{-1}$ cm$^{-1}$ at 560 nm) [42], chrome azurol S ($\epsilon = 1.27 \times 10^5$ L mol$^{-1}$ cm$^{-1}$ at 620 nm) [43], diantipyryl-(2-methoxy)-phenylmethane [44], $N,N$-diethyl-1,4-phenylenediamine ($\epsilon = 5.0 \times 10^4$ L mol$^{-1}$ cm$^{-1}$ at 530 nm) [45], $p$-aminophenylfluorone ($\epsilon = 2.63 \times 10^4$ L mol$^{-1}$ cm$^{-1}$ at 497 nm) [46], 2-hydroxybenzaldiminoglycine [47] and 4-aminoantipyrine (APP) with 1-naphthol ($\epsilon = 2.07 \times 10^4$ L mol$^{-1}$ cm$^{-1}$ at 485 nm) [48]. Most of the methods using aforementioned reagents require heating for colour development, critical pH, and time consuming.

Few indirect spectrophotometric methods for the determination of chromium have been reported in the literature. In these methods, Cr(VI) oxidizes hydroxylamine to nitrite, which is followed by diazotization and coupling reactions. Rathore and Tarafder [49] have used 4-amino phenylthio-acetic acid for diazotization and $N$-(1-naphthyl) ethylenediamine dihydrochloride (NEDA) for coupling reaction. Balasubramanian and Maheswari [50] have proposed $p$-nitroaniline for diazotization and $N$-(1-naphthyl) ethylenediamine dihydrochloride (NEDA) for coupling reaction in acid medium. Revanasiddappa and Kiran Kumar [51] have diazotized the formed nitrite with $p$-aminoacetophenone followed by coupling with citraizinic acid ($\epsilon = 2.12 \times 10^4$ L mol$^{-1}$ cm$^{-1}$ at 470 nm) in an alkaline medium. By a similar reaction, Cherian and Narayana [52] have used $p$-nitroaniline or sulphanilamide and saccharin as diazotizing and coupling reagents, respectively.

Many organic reagents have found application in the extractive spectrophotometric determination of chromium, namely pentamethylene bis-(triphenylphosphonium) bromide ($\epsilon = 1.38 \times 10^3$ at 365 nm) [53], $N$-methylpiperazine-4-carbodithioate ($\epsilon = 5.24 \times 10^3$ at 410 nm) [54], hydroxyamidines ($\epsilon = 1.45 \times 10^4$ L mol$^{-1}$ cm$^{-1}$ at 395 nm) [55], triphenyltetrazolium chloride [56] hydroxyamidines and amides ($\epsilon = 1.18 \times 10^4$ L mol$^{-1}$ cm$^{-1}$ at 408 nm) [57], benzyltributyl ammonium chloride [58], ferron [59], potassium $o,o^'$-dibutylthioephosphate [60], protriptyline hydrochloride [61], 3-(4,5-dimethyl-2-thiazolyl)-2,5-diphenyl-2H--tetrazolium bromide [62] and p-methylisonitrosoacetophenone ($\epsilon = 8.056 \times 10^3$ L mol$^{-1}$ cm$^{-1}$ at 430 nm) [63]. Some of these methods are less sensitive and interfered with several metal ions.

Few extractive spectrophotometric methods available in the literature for chromium determination have been discussed in the following paragraphs. Wasey et al. [64] have extracted the phananthraquinone monoximate of Cr(III) into molten naphthalene-Cr(III) complex, the extract was dissolved in chloroform, and Cr(III) was determined spectrophotometrically at 470 nm against the reagent blank ($\epsilon = 6.65 \times 10^3$ L mol$^{-1}$ cm$^{-1}$). In another method [65], chromium(III) forms 1:3:1 complex with 4-(2-pyridylazo) resorcinol and xylometazoline HCl in acetate buffer of pH 4 to 5.5. This complex was extracted into chloroform and measured the
absorbance of the organic extract at 530 nm \( (\varepsilon = 4.8 \times 10^4 \text{ L mol}^{-1} \text{ cm}^{-1}) \). Beer’s law was obeyed up to 1 \( \mu \text{g mL}^{-1} \) of Cr(III). The method is interfered with several metal ions such as Fe(III), Ni(II), Co(II) and Pd(II). Kamburova \[66\] has reported the 1:1 complexes formed between chromium(VI) and iodonitrotetrazolium chloride, and tetrazolium violet in hydrochloric acid, which were quantitatively extracted into dichloroethane and exhibited absorption maximum at 250 nm \( (\varepsilon = 8.8 \times 10^4 \text{ L mol}^{-1} \text{ cm}^{-1}) \) and at 230 nm \( (\varepsilon = 1.22 \times 10^5 \text{ L mol}^{-1} \text{ cm}^{-1}) \), respectively. The calibration curve was linear up to 0.3 \( \mu \text{g mL}^{-1} \). The same author \[67\] has studied the 1:1 complex formed between Cr(VI) and methylene blue in hydrochloric acid medium. This complex can be extracted into dichloroethane and measured the absorbance at 295 nm \( (\varepsilon = 8.33 \times 10^4 \text{ L mol}^{-1} \text{ cm}^{-1}) \). Both methods have been utilized for the extractive spectrophotometric determination of Cr(VI) in steels and selected soil samples. Few metal ions are interfered. The same author \[68\] has reported nitrotetrazolium blue as an extractive spectrophotometric reagent for the determination of chromium(VI). In this method, Cr(VI)-nitrotetrazolium blue complex was extracted into chloroform and the absorbance of the organic phase was measured at 260 nm \( (\varepsilon = 8.2 \times 10^4 \text{ L mol}^{-1} \text{ cm}^{-1}) \). A calibration graph was linear for 0.01–0.4 \( \mu \text{g mL}^{-1} \) of Cr(VI). Serious interference was caused by W(VI), Hg(II) and perchlorate ions. Burns and Dangolle \[69\] have described a spectrophotometric determination of chromium(VI) by extraction of the dichromate anion into propylene carbonate. The absorbance was measured at 362 nm \( (\varepsilon = 1.95 \times 10^3 \text{ L mol}^{-1} \text{ cm}^{-1}) \). Mn(VI), V(V) and W(VI) interfere seriously. The method was applied to the analysis of chromium in high-speed tool steels. Maheswari and Balasubramanian \[70\] have investigated the ion-pair formation by chromium(VI) with Rhodamine 6G in sulphuric acid medium in the presence of sodium chloride. The ion-pair complex was extracted with toluene. The absorbance of the organic phase was measured at 535 nm \( (\varepsilon = 2.0 \times 10^4 \text{ L mol}^{-1} \text{ cm}^{-1}) \). Beer’s law was obeyed up to 0.8 \( \mu \text{g mL}^{-1} \) of chromium(VI). The method was applied to the analysis of Cr(VI) in steel, geological materials, pharmaceuticals and wastewater.

Recently, Rao et al. \[71\] have presented a spectrophotometric determination of chromium(III) after extraction of its N-methylaniline carbodithioate complex into molten naphthalene. Maximum extraction was obtained in the pH range 2.0-3.5. Naphthalene containing the metal complex was dissolved in DMF and the resulting solution obeys Beer’s law at 340 nm in the concentration range 2.6-31.2 \( \mu \text{g} \) of Cr in 10 mL of the final solution. The method is less sensitive \( (\varepsilon = 8.2 \times 10^3 \text{ L mol}^{-1} \text{ cm}^{-1}) \). Telepckova et al. \[72\] have reported an indirect extractive-spectrophotometric determination of chromium based on the reduction of Cr(VI) to Cr(III) in the
presence of an excess of iodide ion in an acidic medium. The formed triiodide ion reacts with a
cationic dye to form an ion-association complex which was extracted into toluene. The intensely
coloured complex has absorption maximum at 430 nm. The system obeys Beer's law in the
concentration range up to 2 μg mL⁻¹.

A method for non-organic solvent extraction and spectrophotometric determination of
Cr(VI) in alloy steel and Al-alloy was established by Wu and Tan [73]. In HCl-KCl medium
(pH 2.5), Cr(VI)-dibenzylocarbazone complex was formed in acetone solution, and in the presence
of (NH₄)₂SO₄, the complex was extracted by polyethylene glycol 2000 (PEG 2000). The complex
showed an absorption maximum at 545 nm, and apparent molar absorption coefficient was
3.0 × 10⁴ L mol⁻¹ cm⁻¹. Very recently, speciation of Cr(III) and Cr(VI) in environmental samples
by solid phase extraction on Amberson 563 resin has been described [74].

A catalytic spectrophotometric method for the determination of trace Cr(VI) has been
reported [75]. The method was based on the catalytic action of Cr(VI) on the oxidative coupling
reaction of p-amino-N,N-dimethylaniline dihydrochloride and N,N-dimethylaniline in the
presence of H₂O₂. A kinetic spectrophotometric determination of chromium(VI) by oxidation of
sodium pyrogallol-5-sulphonate by hydrogen peroxide was studied by Mitic et al. [76]. Changxiu
Han [77] has proposed a kinetic spectrophotometric method for the determination of Cr(VI) based
on its catalytic effect on the oxidation of methyl orange by potassium bromate. Recently, the
catalytic effect of Cr(III) and Cr(VI) on the oxidation of sulphanilic acid by H₂O₂ was studied
[78]. The reaction was followed spectrophotometrically by measuring the absorbance of the
reaction product at 360 nm. Very recently, Mohamed et al. [79] have developed a kinetic method
for the determination of dissolved chromium species based on the catalytic effect of Cr(III)
and/or Cr(VI) on the oxidation of 2-amino-5-methylphenol (AMP) with H₂O₂.

Some of the sophisticated instrumental techniques which are reported for the analyses of
crchromium include flow-injection spectrophotometric assay [80], sequential-injection
spectrophotometry [81], electrothermal atomic absorption spectrometry [82], spectrofluorimetry
[83], atomic absorption spectrometry[84], GF-AAS [85], voltammetric sensor [86], ion
chromatography inductively coupled plasma mass spectrometry [87], inductively coupled plasma
optical emission spectrometry[88], chemiluminescence [89], stripping voltammetry [90], laser-
induced breakdown spectroscopy [91], X-ray fluorescence analysis [92], mass spectrometry [93]
and gas chromatography-flame photometric detection [94]. The above mentioned techniques
require an expensive experimental set-up and not affordable in most of the laboratories for routine
chemical analysis.
Although numerous methods are available for the determination of chromium, the author has made an attempt to develop simple and cost-effective spectrophotometric methods for the determination of chromium and his findings are presented in this chapter. A direct spectrophotometric determination of chromium with prochlorperazine dimaleate, and an indirect spectrophotometric method for the determination of chromium using p-nitroaniline and citrazinic acid are described in Section 5A and Section 5B, respectively.
**Direct Spectrophotometric Method for the Determination of Chromium with Prochlorperazine Dimaleate**

**5A.1. INTRODUCTION**

The utility of N-alkyl derivatives of phenothiazines as versatile spectrophotometric reagents [95] attracted the author to use prochlorperazine dimaleate (PCPM) for the spectrophotometric determination of chromium. These compounds are first oxidized to a coloured radical cation, which may be further oxidized to a colourless sulphoxide derivative [96]. One of such compounds is PCPM chemically, 2-chloro-10-[3-(4-methyl-1-piperazinyl)propyl]phenothiazine dimaleate, an important piperazine derivative of phenothiazine. It was synthesized by Gritsenko and Zhuravlev [97] using piperazine in butanol, formalin and Raney nickel. It has the following structure [98]:

Prochlorperazine dimaleate (PCPM)

Prochlorperazine dimaleate (PCPM) commercially called as Compazine or Stemetil. It is a white amorphous powder, sparingly soluble in cold water, but soluble in hot water and easily soluble in cold water containing a few drops of hydrochloric acid. It is insoluble in benzene, chloroform and acetone. Its aqueous solution is stable for 5 days at room temperature (28 ± 3°C) and for about 20 days at low temperature (~ 4°C).
5A.2. EXPERIMENTAL

5A.2.1. Instrumentation

ANALYTIC JENA AG model SPECORD-50 and SYSTRONICS-166 digital spectrophotometers with 1.0 cm matched quartz cells were used for all absorbance measurements.

5A.2.2. Reagents

All chemicals used were of analytical reagent grade and doubly distilled water was used throughout.

Standard chromium(VI) solution (1000 µg mL⁻¹): It was prepared by dissolving 0.2829 g K₂Cr₂O₇ in 100 mL distilled water. The stock solution was further diluted as needed.

Standard chromium(III) solution (1000 µg mL⁻¹): This was prepared by dissolving 0.2829 g K₂Cr₂O₇ in 50 mL water, adding 1 mL of saturated sodium sulphite solution, acidifying with 1 mL of 2.5 M sulphuric acid, and then boiling for 2 min to remove excess SO₂ and diluting with water to 100 mL. A suitable volume of this solution was diluted to obtain the working standard.

Prochlorperazine dimaleate (PCPM) (0.5%) (w/v): A 0.5% aqueous solution of PCPM was prepared (insoluble PCPM was dissolved by the addition of few drops of dilute hydrochloric acid) and stored in an amber bottle in a refrigerator.

Other reagents: The following reagents were prepared by dissolving the requisite amount in distilled water; orthophosphoric acid (10 M), bromine water (saturated), sulphosalicylic acid (5%), KOH (4.5 M), sulphuric acid (0.5 and 2.5 M), and HCl (5 M).

5A.2.3. General procedure for the determination of chromium(VI)

Aliquots (0 to 2.0 mL) of the sample solution containing 0 to 20 µg of chromium(VI) were transferred into a series of 10 mL calibrated flasks. Then, volumes of 5 mL of 10 M orthophosphoric acid and 1 mL of 0.5% PCPM solution were added to each flask and the contents were diluted to the mark with distilled water, mixed well and measured the absorbance at 535 nm against the reagent blank. The calibration graph of absorbance against concentration of Cr(VI) is shown in Fig. 5A.1, and it was used for the determination of chromium in the various samples.
5A.2.4. General procedure for the determination of chromium(III)
Aliquots (0 to 2.0 mL) of the sample solution containing 0 to 20 µg of chromium(III) were transferred into a series of 10 mL calibrated flasks. To each, a volume of 0.5 mL of saturated bromine water and 0.5 mL of 4.5 M KOH solution were added, and allowed to stand for 5 min. Then, 0.5 mL each of the 2.5 M sulphuric acid and 5% sulfosalicylic acid were added and followed the above procedure (Section 5A.2.3) for the colour development and the absorbance was recorded at 535 nm against the reagent blank. A calibration curve was drawn and depicted in Fig. 5A.1.

![Absorbance vs. Concentration of Cr(VI)/Cr(III), µg mL⁻¹](image)

**Fig. 5A.1.** Beer’s law plot of Cr(VI) - PCPM system

5A.2.5. Analysis of mixture containing chromium(III) and chromium(VI)
An aliquot (20 µg) of the mixture was analyzed according to the procedure in the Section 5A.2.3, to establish the concentration of chromium(VI). Another aliquot (20 µg) of the mixture was analyzed according to the procedure in the Section 5A.2.4, to establish the concentration of total chromium [Cr(III) + Cr(VI)]. The concentration of chromium(III) in the mixture was calculated by subtracting the concentration of chromium(VI) from the concentration of total chromium [Cr(III) + Cr(VI)].
5A.2.6. Analysis of chromium in alloy steels
Alloy steels [GKW Steel, India (0.05 g) and Stainless Steel No. 304 (0.05 g)] were dissolved in approximately 8 mL of aqua-regia and evaporated nearly to dryness on a sand bath. Sulphuric acid (1-2 mL, 1:1) was then added and evaporated until salts crystallized, cooled and 10 mL of water was added. The solution was warmed, filtered, and treated with 10 mL of 5 M HCl and 10 mL of MIBK to extract iron [67,99]. The aqueous layer was separated and diluted to a known volume with water. Suitable aliquots of sample solutions were analyzed according to the procedure for chromium(III) [Section 5A.2.4].

5A.2.7. Analysis of chromium in industrial effluents
The samples of chromium plating effluent and tannery effluent were diluted 10 and 40 times, respectively before the analysis. Suitable aliquot of sample solutions were analyzed according to the procedure for determining chromium(VI) [Section 5A.2.3] and chromium(III) [Section 5A.2.4]. A parallel determination was carried out according to the standard diphenylcarbazide method [6,8].

5A.2.8. Analysis of chromium in soil
A known amount of (2 g) air dried homogenized soil sample was fused with anhydrous sodium carbonate [100] in a silica crucible and evaporated to dryness after the addition of 25 mL water. The dried material was dissolved in water, filtered through Whatman No. 40 filter paper. The filtrate was diluted to a known volume with water (100 mL). An aliquot of this sample solution was analyzed for chromium according to general procedure for chromium(III) [Section 5A.2.4]. Sample solutions were also analyzed according to the standard diphenylcarbazide method [6,8].

5A.2.9. Analysis of chromium in water
Each filtered environmental water sample (100 mL) was analyzed for chromium. They tested negative. To these samples known amounts of chromium(VI) (400 µg) were spiked and analyzed for chromium by the proposed method and by the standard diphenylcarbazide method in order to check the validity of the developed method.
5A.3. RESULTS AND DISCUSSION

In this investigation, chromium(VI) oxidize PCPM instantaneously at room temperature (28 ± 3°C) in orthophosphoric acid medium to form a red-coloured species [96]. PCPM undergo one-electron reversible oxidation to a stable red-coloured intermediate, which is believed to be a radical cation [101 - 103] with a maximum absorption at 535 nm. The possible reaction pathway between PCPM and Cr(VI) is presented in Scheme 5A.1. The formation of radical cation was confirmed by the ion-exchange technique.

\[
\begin{align*}
\text{PCPM} &\quad \text{Cr(VI)} \\
\begin{array}{c}
\text{N} \\
\text{R} \\
\text{Cl}
\end{array} &\quad + e^- \\
\text{N} &\quad + e^- \\
\begin{array}{c}
\text{N} \\
\text{R} \\
\text{Cl}
\end{array}
\end{align*}
\]

Scheme 5A.1. Proposed reaction pathway

5A.3.1. Nature of the coloured species

A solution of the coloured species formed between PCPM–chromium(VI) was passed through a Dowex 50W–X8 cation-exchange column (26 × 1.5 cm), and Dowex 1–X8 anion-exchange resin. The electrolytic nature of the species was established by the retention on the cationic or anionic resin. The colour was retained on Dowex 50W–X8 and not on Dowex 1–X8, anion exchange resin. This confirmed that the coloured species was cationic.

5A.3.2. Absorption spectra

A known amount of the analyte [Cr(VI), 1.0 µg mL⁻¹] was taken and the general procedure for the determination of chromium(VI) [Section 5A.2.3] was followed for the colour development, and the absorbance of the red coloured species was recorded against the reagent blank in the wavelength range 400 – 670 nm. The absorption spectra of the red coloured species and the
reagent blank are shown in Fig 5A.2. The red species exhibits maximum absorption at 535 nm. The reagent blank had negligible absorbance at this wavelength.

![Absorption spectra graph](image)

**Fig. 5A.2.** Absorption spectra of (a) Cr(VI)–PCPM coloured species [Cr(VI), 1.0 μg mL⁻¹] vs. reagent blank and (b) reagent blank vs. distilled water.

### 5A.3.3. Optimization of experimental parameters

The experimental conditions were optimized at 535 nm by studying the influence of following parameters with 10 μg of chromoium(VI) in a final volume of 10 mL.

#### 5A.3.3.1. Effects of the acids and the acid concentration

The sensitivity and stability of the red species depends upon the nature and the concentration of the acid used. The reaction was tested in different acid medium (hydrochloric, sulphuric and orthophosphoric acid). The red-colour was found to be unstable in hydrochloric acid and sulphuric acid medium. Orthophosphoric acid medium is recommended because the red colour was stable for 2 h and subject to less interference from foreign ions. The effect of varying concentration of orthophosphoric acid on the absorbance of the red species was studied. The maximum colour development obtained in the range 3-7 M orthophosphoric acid, and hence an overall acid strength of 5 M (5 mL of 10 M orthophosphoric acid in a final volume of 10 mL) was
selected and thus used in subsequent studies. At lower concentrations of phosphoric acid (< 3.0 M), the reaction was not only less sensitive but also required more time for complete colour development. Higher acid concentration is required because the hydrogen ions stabilize the semiquinonoid free radical by decreasing the rate of its disproportionation and subsequent hydrolysis [96], which may occur according to the following reactions.

\[
\begin{align*}
2 & \quad \begin{array}{c}
\text{S} \\
\text{N} \\
\text{R} \\
\text{Cl}
\end{array} & \quad \rightarrow & \quad \begin{array}{c}
\text{S} \\
\text{N} \\
\text{R} \\
\text{Cl}
\end{array} & \quad + & \quad \begin{array}{c}
\text{S} \\
\text{N} \\
\text{R} \\
\text{Cl}
\end{array} & \quad + & \quad \text{H}^+ \\
\text{(Coloured radical cation)}
\end{align*}
\]

\[
\begin{align*}
\text{S} \\
\text{N} \\
\text{R} \\
\text{Cl}
\end{array} & \quad + & \quad \text{H}_2\text{O} & \quad \rightarrow & \quad \begin{array}{c}
\text{S} \\
\text{N} \\
\text{R} \\
\text{Cl}
\end{array} & \quad + & \quad \text{H}^+ \\
\text{(Colourless sulfoxide)}
\end{align*}
\]

\[
R = -(\text{CH}_2)_5\text{N} \begin{array}{c}
\text{N} \\
\text{CH}_3
\end{array} \cdot 2\text{C}_4\text{H}_4\text{O}_4
\]

54.3.2. Effect of the reagent concentration
The effect of PCPM concentration on absorbance of the coloured system was examined at 535 nm by keeping a fixed concentration of chromium(VI) [1 \(\mu\)g mL\(^{-1}\)] and varying the volume of PCPM (0.5%). The absorbance values are found to be optimum in the range 0.5 – 1.5 mL of 0.5% PCPM (Fig. 5A.3). A volume of 1.0 mL of 0.5% PCPM in a total volume of 10 mL was taken for further investigation.
5A.3.3. Effects of the temperature and time
The reaction was carried out at room temperature (28 ± 3 °C) and the temperature in the range 5 – 50 °C had no effect on the colour system. A decrease in absorbance was observed at higher temperature.

The red-coloured radical cation was formed rapidly under the optimum experimental conditions and it was found to be stable for 2 h at room temperature.

5A.3.4. Choice of oxidizing agent
Chromium(III) was determined [Section 4A.2.4] after its oxidation to chromium(VI). Various oxidizing agents [8] such as, persulphate in the presence of a silver ion as the catalyst in an acid solution, permanganate in an acid medium, and hydrogen peroxide and bromine in an alkaline medium have been used. For all these methods, excess oxidizing agent must be destroyed before the determination of chromium(VI) to avoid the interference with the determination. Usually, excess persulphate and peroxide were removed by boiling, and adding sodium azide destroyed permanganate. In the present investigation, bromine water in an alkaline medium was used to oxidize Cr(III) to Cr(VI), and sulphosalicylic acid (5%) was used to remove the excess of bromine.

Fig. 5A.3. Effect of PCPM (0.5%) concentration on the absorbance of red-coloured radical cation [Cr(VI), 1 µg mL⁻¹]
5A.3.5. Interference studies

In order to evaluate the suitability of the proposed method, the effect of various ions which are likely to interfere in the determination of chromium(VI) was studied by adding a known amount of diverse ions to 1 µg mL⁻¹ chromium(VI) solution. The tolerance limits of interfering species were established at those concentrations that do not cause more than ±2% error in absorbance values. The tolerance limits of foreign ions are listed in Table 5.1. The results indicate that Fe(II), iodide and iodate, susceptible in the determination of Cr(VI). However, interference of foreign ions could be obviated by using appropriate masking agents in the analysis of samples. The interference of vanadium(V) and cerium(IV) can be overcome by extraction of chromium(VI) as chromyl chloride in 5 mL MIBK after addition of 5 M HCl to provide an overall acidity of 0.3–0.5 M. Chromium(VI) in the organic layer can be stripped by equilibration with 5 mL of water for determination. The major advantage of this method is that PCPM can be used as a selective reagent for the determination of chromium in the presence of large amounts of Fe(III), Mo(VI), Ni(II) and Mn(II).

Table 5.1. Effect of interfering ions on the determination of Cr(VI) [1 µg mL⁻¹ ].

<table>
<thead>
<tr>
<th>Interferents</th>
<th>Tolerance limit (µg mL⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ba²⁺, EDTA</td>
<td>≥ 4000</td>
</tr>
<tr>
<td>Al³⁺, Cu²⁺, Mn²⁺, Ni²⁺, Zn²⁺, Ca²⁺, NO₃⁻, Br⁻, Cl⁻, acetate, citrate, oxalate</td>
<td>≥ 1000</td>
</tr>
<tr>
<td>Fe⁶⁺, MoO₄²⁻, F, tartrate</td>
<td>≤ 700</td>
</tr>
<tr>
<td>Co³⁺, Bi³⁺, WO₄²⁻</td>
<td>≤ 100</td>
</tr>
<tr>
<td>Fe²⁺, Ce⁴⁺, I⁻, iodate, VO₃⁻</td>
<td>≤ 1</td>
</tr>
</tbody>
</table>
5A.3.6. Optical characteristics and analytical data
A linear calibration graph was obtained for 2–20 μg of chromium(VI) in the final volume of 10 mL. The correlation coefficient of the calibration graph equals 0.9999 [y = (0.396)x + 0.003; where ‘x’ is the concentration of chromium(VI) in μg mL⁻¹]. The molar absorption coefficient (ε), specific absorptivity (α) and Sandell’s sensitivity (S) of the method were found to be 2.09 × 10⁴ L mol⁻¹ cm⁻¹, 0.4 mL g⁻¹ cm⁻¹, and 2.49 ng cm⁻², respectively. The detection limit (DL = 3.3 σ/S) and quantitation limit (QL = 10 σ/S) [where ‘σ’ the standard deviation of reagent blank (n = 10) and ‘S’ is the slope of calibration curve] of chromium(VI) determination were found to be 40 and 120 ng mL⁻¹, respectively. The precision of the method was established by analysis of standard solutions of 6, 10 and 14 μg chromium(VI) in a final volume of 10 mL. Ten replicate determinations of each concentration gave relative standard deviations (RSD) of 0.25, 0.1 and 0.06%, respectively.

5A.3.7. Applications
The proposed procedure has been successfully applied to analyze chromium in alloy steels, industrial effluents, soil and water [Sections 5A.2.6 to 5A.2.9]. Parallel determinations were carried out for the determination of chromium in the above sample solutions with the standard diphenylcarbazide (DPC) method [6,8]. It involves the reaction of Cr(VI) with DPC in sulphuric acid medium to give violet-coloured solution (λᵥₐₓ = 545 nm). The results are tabulated in Tables 5.2 to 5.5.

5A.3.8. Statistical evaluation of the results
In order to validate the results of the proposed method, the certified values and the results of the standard method were statistically evaluated with the results of proposed method by F- and t-tests at 95% confidence level. The F- and t- values [Tables 5.2 to 5.5] reveal the accuracy of the method.
Table 5.2. Determination of chromium in alloy steels

<table>
<thead>
<tr>
<th>Sample (w/v)</th>
<th>Certified chromium (%)</th>
<th>Volume of solution (mL)</th>
<th>Chromium found (%)(^a)</th>
<th>Recovery (%)</th>
<th>RSD (%)</th>
<th>t-test(^b)</th>
</tr>
</thead>
<tbody>
<tr>
<td>GKW steel, India [C 0.54%, Mn 0.89%, S 0.018%, P 0.034%, Si 0.33%, V 0.13%]</td>
<td>1.02</td>
<td>1.0</td>
<td>1.010 ± 0.02</td>
<td>99.0</td>
<td>1.98</td>
<td>1.12</td>
</tr>
<tr>
<td>Stainless Steel No. 304 [Ni 8.12%, Fe (70-71%)]</td>
<td>18.0</td>
<td>1.0</td>
<td>17.90 ± 0.09</td>
<td>99.4</td>
<td>0.50</td>
<td>2.50</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1.5</td>
<td>17.91 ± 0.08</td>
<td>99.5</td>
<td>0.45</td>
<td>2.52</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2.0</td>
<td>17.92 ± 0.07</td>
<td>99.6</td>
<td>0.40</td>
<td>2.56</td>
</tr>
</tbody>
</table>

\(^a\) Mean ± standard deviation (n = 5).

\(^b\) Tabulated t-value for 4 degrees of freedom at P (0.95) is 2.776.

\(^c\) Solution diluted 20 times before determination.

Table 5.3. Determination of chromium in industrial effluents

<table>
<thead>
<tr>
<th>Sample</th>
<th>Chromium found (µg ml(^{-1}))(^a)</th>
<th>F-test(^b)</th>
<th>t-test(^c)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Proposed method</td>
<td>Reference method</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Cr(III)</td>
<td>Cr(VI)</td>
<td>Cr(III)</td>
</tr>
<tr>
<td>Chromium plating effluent(^d)</td>
<td>28.0±0.5</td>
<td>63.9±0.3</td>
<td>28.1±0.6</td>
</tr>
<tr>
<td>Tannery effluent(^e)</td>
<td>258.0±0.4</td>
<td>ND(^f)</td>
<td>258.2±0.5</td>
</tr>
</tbody>
</table>

\(^a\) Mean ± standard deviation (n = 5).

\(^b\) Tabulated F-value for (4,4) degrees of freedom at P (0.95) is 6.39.

\(^c\) Tabulated t-value for 8 degrees of freedom at P (0.95) is 2.306.

\(^d\) Solution diluted 10 times before determination.

\(^e\) Solution diluted 40 times before determination.

\(^f\) ND, Not detected.
Table 5.4. Determination of chromium in soil

<table>
<thead>
<tr>
<th>Sample (w/v)</th>
<th>Proposed method</th>
<th>Reference method [6,8]</th>
<th>F-test</th>
<th>t-test</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Chromium found (µg g⁻¹)</td>
<td>RSD</td>
<td>Chromium found (µg g⁻¹)</td>
<td>RSD</td>
</tr>
<tr>
<td>Soil-1 (2 g/100 mL)</td>
<td>183.2±0.8</td>
<td>0.44</td>
<td>183.9±0.9</td>
<td>0.49</td>
</tr>
<tr>
<td>Soil-2 (2 g/100 mL)</td>
<td>213.5±0.7</td>
<td>0.33</td>
<td>214.4±0.9</td>
<td>0.42</td>
</tr>
</tbody>
</table>

a Mean ± standard deviation (n = 5).
b Tabulated F-value for (4,4) degrees of freedom at P (0.95) is 6.39.
c Tabulated t-value for 8 degrees of freedom at P (0.95) is 2.306.

Table 5.5. Determination of chromium in water samples

<table>
<thead>
<tr>
<th>Sample</th>
<th>Cr taken (µg)</th>
<th>Proposed method</th>
<th>Reference method [6,8]</th>
<th>F-test</th>
<th>t-test</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Cr found (µg)</td>
<td>Recovery (%)</td>
<td>RSD (%)</td>
<td>Cr found (µg)</td>
<td>Recovery (%)</td>
</tr>
<tr>
<td>River water</td>
<td>4.0</td>
<td>3.98±0.04</td>
<td>99.5</td>
<td>1.0</td>
<td>3.98±0.05</td>
</tr>
<tr>
<td></td>
<td>8.0</td>
<td>7.97±0.03</td>
<td>99.6</td>
<td>0.4</td>
<td>7.98±0.04</td>
</tr>
<tr>
<td>Lake water</td>
<td>4.0</td>
<td>3.96±0.05</td>
<td>99.0</td>
<td>1.3</td>
<td>3.97±0.06</td>
</tr>
<tr>
<td></td>
<td>8.0</td>
<td>7.94±0.03</td>
<td>99.3</td>
<td>0.4</td>
<td>7.95±0.05</td>
</tr>
</tbody>
</table>

a Mean ± standard deviation (n = 5).
b Tabulated F-value for (4,4) degrees of freedom at P (0.95) is 6.39.
c Tabulated t-value for 8 degrees of freedom at P (0.95) is 2.306.

5A.4. CONCLUSIONS

The proposed method offers a simple, rapid and accurate and reproducible procedure for the determination of chromium. It requires no heating or tedious extraction steps. Thus, the use of organic solvents, which are generally toxic, is avoided. The developed method has several advantages, the major advantages being stability (2 h) [30 min in the standard DPC method] and sensitivity of the colour system. The results obtained by the proposed and the standard diphenylcarbazide methods are in good agreement. The method has the added advantage of determining individual amounts of chromium(VI) and chromium(III).
Indirect Spectrophotometric Method for the Determination of Chromium Using \( p \)-Nitroaniline and Citrazinic acid

5B.1. INTRODUCTION
Citrazinic acid (CZA), chemically 2,6-dihydroxyisonicotinic acid or 1-2-dihydro-6-hydroxy-2-oxopyridine-4-carboxylic acid. It is yellowish powder with a greenish tinge. It is insoluble in water and slightly soluble in hot hydrochloric acid. CZA is freely soluble in alkali hydroxide or carbonate solutions. Alkaline solutions turn blue on standing. It has the following structure:

![CZA](image)

Kaventitis has used citrazinic acid as a reagent in the spectrophotometric determination of uranium(VI) and iron(III) [104], and kinetic spectrophotometric determination of iron(III), copper(II) and vanadium(V) [105]. Revanasiddappa and Kiran Kumar [51] have used CZA as a coupling agent with \( p \)-aminoacetophenone for the determination of chromium. The same authors have used CZA as a coupling agent for the determination of nitrite with \( p \)-aminoacetophenone [106] and \( p \)-nitroaniline [107]. It has also been found application as coupling agent in the assay of some chemotherapeutic agents by spectrophotometry [108]. In this section, the author has presented his research investigation of indirect spectrophotometric method for the determination of chromium using \( p \)-nitroaniline as a diazotizing reagent and citrazinic acid as a coupling agent, and its application in the determination of chromium in alloy steels, industrial effluents, soil and water.
5B.2. EXPERIMENTAL

5B.2.1. Instrumentation
ANALYTIC JENA AG model SPECORD-50 and an ELICO model SL–171 digital spectrophotometers with 1.0 cm matched quartz cells were used for all absorbance measurements. The pH measurements were made with an ELICO (model LI-610) digital pH meter.

5B.2.2. Reagents
All chemicals used were of analytical reagent grade and doubly distilled water was used throughout.

Preparations of standard solutions of chromium(VI) and chromium(III) are described in Section 5A.2.2.

*p-Nitroaniline (PNA) (0.05%):* This was prepared by dissolving 0.125 g *p*-nitroaniline in 55.3 mL concentrated hydrochloric acid and diluting to 250 mL with water. The resulting solution was 0.05% PNA in 2.5 M HCl.

_Citrazinic acid (CZA) (0.1%):*_ It was prepared by dissolving 0.1 g of citrazinic acid in 2 mL of 4 M NaOH and diluting to 100 mL with water.

_Acetate buffer (pH 4.0):*_ It was prepared by dissolving 13.6 g of sodium acetate trihydrate in 80 mL water. Solution pH was adjusted to 4.0 with acetic acid, and the mixture was diluted to 100 mL with water.

_Others:_ Hydroxylamine hydrochloride (0.1%), bromine water (saturated), sulphosalicylic acid (5%), KOH (4.5 M), sulphuric acid (0.5 M and 2.5 M), HCl (5 M), NaOH (4 M) and methyl isobutyl ketone (MIBK) were used.

5B.2.3. General procedure for the determination of chromium(VI)
Aliquots (0 to 1.8 mL) of the standard solution containing 0-18 µg of chromium(VI) was transferred to a series of 10 mL of calibrated flasks. Then, volumes of 0.5 mL each of the acetate buffer (pH 4.0) and 0.1% hydroxylamine hydrochloride solution were added. After 2 min, a volume of 0.5 mL of 0.05% *p*-nitroaniline (dissolved in 2.5 M HCl) was added and allowed to stand for two min. Then, volumes of 2 mL each of the 0.1% citrazinic acid and 4 M sodium hydroxide were added, and the mixture was diluted to 10 mL with distilled water and mixed well. After 5 min, the absorbance of the coloured azo dye was measured at 515 nm against the reagent
blank prepared in the same manner but containing no chromium(VI). The calibration curve is depicted in Fig. 5B.1.

![Absorbance vs Concentration Graph](image)

**Fig. 5B.1.** Beer’s law plot of Cr(VI)–PNA–CZA system

### 5B.2.4. General procedure for the determination of chromium(III)

Aliquots of the standard solution containing 2-18 µg of chromium(III) was transferred into a series of 10 mL calibrated flasks. To each, a volume of 0.5 mL of saturated bromine water and 0.5 mL of 4.5 M KOH solution were added and allowed to stand for 5 min. Then, 0.5 mL each of the 2.5 M sulphuric acid and 5% sulphosalicylic acid were added and developed an azo dye by following the above procedure (Section 5B.2.3). The absorbance of the coloured azo dye was recorded at 515 nm against the reagent blank, omitting chromium(III). A calibration graph was drawn (Fig. 5B.1) and this graph was used to determine Cr(III) content in the samples solution.

### 5B.2.5. Determination of chromium in the mixture containing chromium(III) and chromium(VI)

An aliquot (18 µg) of the mixture was analyzed according to the procedure in the Section 5B.2.3, to establish the concentration of chromium(VI). Another aliquot (18 µg) of the mixture was analyzed according to the procedure the Section 5B.2.4, to establish the concentration of total chromium [Cr(III) + Cr(VI)]. The concentration of chromium(III) in the mixture was calculated
by subtracting the concentration of chromium(VI) from the concentration of total chromium \([\text{Cr(III)} + \text{Cr(VI)}]\).

**5B.2.6. Determination of chromium in alloy steels, industrial effluents, soil and water samples**

The detailed procedures for the analysis of the above samples are described in Sections 5A.2.6 to 5A.2.9. The chromium content was then determined in the samples by following the general procedures for Cr(VI) and Cr(III) which are presented in the Sections 5B.2.4. and 5B.2.5.

**5B.3. RESULTS AND DISCUSSION**

In this method, chromium(VI) oxidizes hydroxylamine to generate nitrite, which was used to diazotize \(p\)-nitroaniline (PNA). The formed diazonium salt was then coupled with citrazinic acid (CZA) in an alkaline medium to give a red-coloured azo dye, which showed a maximum absorbance at 515 nm.

In the preliminary investigation, an amount of 10 \(\mu g\) chromium(VI) and 0.1% hydroxylamine hydrochloride were used. The proposed reaction scheme (Scheme 5B.1) shows that 10 \(\mu g\) chromium(VI) reacts with hydroxylamine to generate 6.6 \(\mu g\) nitrite. Quantitative oxidation of hydroxylamine to nitrite by chromium(VI) was confirmed by performing the diazo-coupling reaction with standard nitrite solution, using CZA as the coupling agent. Ten micrograms of chromium(VI), on treatment with hydroxylamine and after the diazo-coupling reaction in total volume of 10 mL gives an absorbance of 0.40 \(\pm\) 0.01 A, at 515 nm, comparable with 0.40 A, the absorbance at 515 nm from the diazo-coupling reaction of 6.6 \(\mu g\) of standard nitrite solution in a total aqueous volume of 10 mL. This confirms that 10 \(\mu g\) of Cr(VI) generated 6.6 \(\mu g\) of nitrite when it reacts with 0.5 mL of 0.1% hydroxylamine hydrochloride in an acetate buffer (pH 4.0).
Scheme 5B.1. Proposed reaction pathway

5B.3.1. Absorption spectra

The general procedure for the determination of Cr(VI) [Section 5B.2.3] was followed for the formation of azo dye [Cr(VI), 1 µg mL⁻¹] and the wavelength of maximum absorption was determined by recording the absorbance of the azo dye in the wavelength range 400 - 650 nm.
The absorbance of the reagent blank against distilled water was also scanned in the same wavelength range. Absorption spectra [Fig. 5B.2] were obtained by plotting the absorbance values against wavelength. The azo dye showed maximum absorption at 515 nm.

**Fig. 5B.2.** Absorption spectra of (a) the azo dye [Cr(VI), 1 µg mL\(^{-1}\)] vs. reagent blank and (b) reagent blank vs. distilled water.

**5B.3.2. Optimization of experimental parameters**

In order to establish the linear range for quantitative determination of chromium in various samples, the following experimental variables were studied at 515 nm with 1 µg mL\(^{-1}\) of Cr(VI).

**5B.3.2.1. Effect of pH**

The oxidation of hydroxylamine to nitrite by chromium(VI) was effective in the pH range 3.5 – 4.5. A change in this pH range resulted in decreased absorbance of the azo dye. Hence, the above pH range was maintained by adding 0.5 mL acetate buffer (pH 4.0). This reaction is similar to oxidation of hydroxylamine to nitrite by iodine [109,110].

**5B.3.2.2. Effects of the acid and PNA concentration on diazotization**

The effect of acidity on the diazotization reaction was studied in the range 0.025 - 0.25 M HCl in a final volume of 10 mL. A constant and maximum absorbance of the azo dye was observed in an overall acidity of 0.05 - 0.2 M HCl. A decrease in absorbance was noticed above or below this
range. Hence, an overall acidity of 0.125 M HCl for the diazotization was maintained by the addition of 0.5 mL of 0.05% PNA in a total volume of 10 mL. The effect of PNA concentration on the absorbance of the azo-dye was studied and presented in Fig. 5B.3.

![Graph showing the effect of PNA on absorbance]

**Fig. 5B.3.** Effect of PNA (0.05%) on absorbance of the azo dye, [Cr(VI), 1 µg mL⁻¹]

### 5B.3.2.3. Effect of CZA concentration

The influence of CZA concentration on the colour intensity of the azo dye is depicted in Fig. 5B.4. The results indicate that the constant absorbance values were obtained in the range

![Graph showing the effect of CZA on absorbance]

**Fig. 5B.4.** Effect of CZA (0.1%) on absorbance of the azo dye, [Cr(VI), 1 µg mL⁻¹]
1.5 - 2.5 mL of 0.1% CZA. A slight decrease in absorbance of the azo dye was observed at higher concentrations of CZA. Therefore, further investigations were performed with 2 mL of 0.1% CZA in a final volume of 10 mL.

5B.3.2.4. Effect of NaOH concentration
The stability and formation of an azo-dye depends upon the nature of reaction medium. An alkaline medium is the best for this. In this system, sodium hydroxide, sodium carbonate and aqueous ammonia were investigated, but sodium hydroxide was found to be more suitable for the coupling reaction compared to sodium carbonate or aqueous ammonia because the formed dye was stable and more intense in sodium hydroxide medium. The dye was unstable in presence of aqueous ammonia, and less intense in sodium carbonate solution. A maximum and constant absorbance values were obtained from 1.0 to 4.0 mL of 4 M NaOH, and a volume of 2.0 mL of 4 M NaOH was selected and thus used. The results of effect of NaOH concentration on the absorbance of the azo dye is graphically presented in Fig. 5B.5.

![Graph showing effect of NaOH concentration on absorbance](image)

**Fig. 5B.5.** Effect of NaOH (4 M) on the absorbance of the azo dye, [Cr(VI), 1 µg mL⁻¹]

5B.3.2.5. Effects of the temperature and time
The oxidation of hydroxylamine to nitrite by chromium(VI) was completed within 2 min at room temperature (27 ± 3 °C). The diazotization was also carried out at room temperature, and 2 min was the minimum time required for the diazotization. No cooling (<5 °C) was required for the diazotization. A time of 5 min was required for completion of coupling reaction. The formed azo dye was stable for 4 h.
5B.3.3. Choice of oxidizing agent

Oxidation of Cr(III) to Cr(VI) has been discussed in Section 5A.3.4. The present method has also utilized bromine water in an alkaline medium to oxidize Cr(III) to Cr(VI), and sulphosalicylic acid (5%) was used to remove the unreacted bromine.

5B.3.4. Interference studies

The effect of various ions which are likely to interfere in the determination of chromium(VI) was studied by adding a known amount of diverse ions to 1 μg mL⁻¹ chromium(VI) solution. The tolerance limits of interfering species were established at those concentrations that do not cause more than ± 2% error in absorbance values. Ce(IV), Cu(II), Mn(VII), Ge(IV), Fe(II), Sn(II), Sb(III), V(V) and W(VI) caused negative interference. Fe(III) showed positive interference. The results indicate that Cu(II), Fe(II), Fe(III) and Co(II) are highly susceptible in the determination of Cr(VI). The interference of Cu(II), Fe(III) and Co(II) can be masked by addition of 1 mL of 1% EDTA. The interference of V(V), Mn(VII) and Ce(IV) can be overcome by extraction of chromium(VI) as chromyl chloride in 5 mL MIBK after addition of 5 M HCl to provide an overall acidity of 0.3-0.5 M [111]. Chromium(VI) in the organic layer can be stripped by equilibration with 5 mL of water for determination. The interference of other foreign ions can be obviated by using appropriate masking agents in the analysis of samples. The tolerance limit of foreign ions is listed in Table 5.6.

<table>
<thead>
<tr>
<th>Interferents</th>
<th>Tolerance limit (μg mL⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Na⁺, F⁻, SO₄²⁻, CH₃COO⁻, EDTA</td>
<td>≥ 3000</td>
</tr>
<tr>
<td>Mg²⁺, IO₃⁻, AsO₃³⁻, PO₄³⁻, oxalate, tartrate</td>
<td>≥ 2000</td>
</tr>
<tr>
<td>Bi⁷⁺, MoO₄²⁻, citrate</td>
<td>≤ 1500</td>
</tr>
<tr>
<td>Pb⁺⁺, Cd⁺⁺, Mn⁺⁺, Zn⁺⁺</td>
<td>≤ 500</td>
</tr>
<tr>
<td>Ni²⁺, Ag⁺, Hg²⁺, Ce³⁺, Ge⁴⁺, Sb³⁺, Sn⁺⁺, MnO₄⁻, WO₄²⁻</td>
<td>≤ 100</td>
</tr>
<tr>
<td>Fe⁺⁺, Fe⁺⁺, Cu⁺⁺, Co⁺⁺, VO₃⁻</td>
<td>≤ 10</td>
</tr>
</tbody>
</table>

Table 5.6. Effect of interfering ions on the determination of chromium(VI) [1 μg mL⁻¹]
5B.3.5. Optical characteristics and analytical data

A linear calibration graph was obtained for 2 - 18 µg of chromium(VI) in a final volume of 10 mL. The slope and intercept were 0.4004 and 0.0015, respectively \[y = (0.4004)x + 0.0015; \] where \(x\) is the concentration of Cr(VI) in µg mL\(^{-1}\)]. The calibration graph has a correlation coefficient of 0.999. The detection limit (DL = 3.3 \(\sigma\)/S) and quantitation limit (QL = 10 \(\sigma\)/S), where \(\sigma\) is the standard deviation of reagent blank (n=10) and \(S\) is the slope of calibration curve] of chromium determination were found to be 50 and 150 ng mL\(^{-1}\), respectively. The molar absorptivity (\(\varepsilon\)) and Sandell’s sensitivity (S) of the proposed method were \(2.1 \times 10^4\) L mol\(^{-1}\) cm\(^{-1}\) and 2.5 ng cm\(^{-2}\), respectively. The reproducibility of the method was established by an analysis of standard solutions of 2, 6 and 10 µg of chromium in a final volume of 10 mL with relative standard deviations (RSD) (n = 10) of 0.3, 0.09 and 0.06%, respectively.

5B.3.6. Applications

The described procedure was satisfactorily applied to the determination of chromium in alloy steels, industrial effluents, water and soil samples [Section 5B.2.6]. Parallel determinations were also carried out with the standard diphenylcarbazide (DPC) method [6,8]. The results are presented in Tables 5.7 to 5.9. The results of the proposed and standard methods are subjected to statistical evaluation by the Student’s t-test and F-test at 95% confidence level. The statistical data indicates the validity of the developed method.

<table>
<thead>
<tr>
<th>Sample (w/v)</th>
<th>Certified chromium (%)</th>
<th>Volume of solution (mL)</th>
<th>Chromium found (%) (^a)</th>
<th>Recovery (%)</th>
<th>RSD (%)</th>
<th>t-test (^b)</th>
</tr>
</thead>
<tbody>
<tr>
<td>GKW steel, India (0.05 g/100 mL) [C 0.54%, Mn 0.89%, S 0.018%, P 0.034%, Si 0.33%, V 0.13%]</td>
<td>1.02</td>
<td>1.0</td>
<td>1.000±0.02</td>
<td>98.0</td>
<td>2.0</td>
<td>2.2</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1.5</td>
<td>1.010±0.01</td>
<td>99.0</td>
<td>1.0</td>
<td>2.2</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2.0</td>
<td>1.012±0.01</td>
<td>99.2</td>
<td>1.0</td>
<td>1.8</td>
</tr>
<tr>
<td>Stainless Steel No. 304 (0.05 g/100 mL) (^c) [Ni 8.12%, Fe (70-71%)]</td>
<td>18.0</td>
<td>1.0</td>
<td>17.80±0.18</td>
<td>98.9</td>
<td>0.67</td>
<td>2.48</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1.5</td>
<td>17.90±0.15</td>
<td>99.4</td>
<td>0.61</td>
<td>1.49</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2.0</td>
<td>17.91±0.14</td>
<td>99.5</td>
<td>0.56</td>
<td>1.44</td>
</tr>
</tbody>
</table>

\(^a\) Mean ± standard deviation (n = 5).
\(^b\) Tabulated t-value for 4 degrees of freedom at \(P (0.95)\) is 2.776.
\(^c\) Solution diluted 20 times before determination.
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**Table 5.8.** Determination of chromium in industrial effluents

<table>
<thead>
<tr>
<th>Sample</th>
<th>Chromium found (µg mL⁻¹)²</th>
<th>F-test b</th>
<th>t-test c</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Cr(III)</td>
<td>Cr(VI)</td>
<td>Cr(III)</td>
</tr>
<tr>
<td>Chromium plating effluent d</td>
<td>27.8±0.6</td>
<td>63.7±0.4</td>
<td>27.9±0.7</td>
</tr>
<tr>
<td>Tannery effluent e</td>
<td>257.5±0.5</td>
<td>ND f</td>
<td>257.7±0.6</td>
</tr>
</tbody>
</table>

³ Mean ± standard deviation (n = 5).
⁴ Tabulated F-value for (4,4) degrees of freedom at P (0.95) is 6.39.
⁵ Tabulated t-value for 8 degrees of freedom at P (0.95) is 2.306.
⁶ Solution diluted 10 times before determination.
⁷ Solution diluted 40 times before determination.
⁸ ND, Not detected.

---

**Table 5.9.** Determination of chromium in water and soil samples

<table>
<thead>
<tr>
<th>Sample</th>
<th>Cr taken (µg)</th>
<th>Proposed method</th>
<th>Reference method [6,8]</th>
<th>F-test b</th>
<th>t-test c</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Cr found (µg)</td>
<td>Recovery (%)</td>
<td>RSD (%)</td>
<td>Cr found (µg)</td>
<td>Recovery (%)</td>
</tr>
<tr>
<td>River water</td>
<td>4.0</td>
<td>3.97±0.05</td>
<td>99.3</td>
<td>1.3</td>
<td>3.98±0.06</td>
</tr>
<tr>
<td></td>
<td>8.0</td>
<td>7.96±0.03</td>
<td>99.5</td>
<td>0.4</td>
<td>7.97±0.04</td>
</tr>
<tr>
<td>Lake water</td>
<td>4.0</td>
<td>3.96±0.06</td>
<td>99.0</td>
<td>1.5</td>
<td>3.97±0.07</td>
</tr>
<tr>
<td></td>
<td>8.0</td>
<td>7.95±0.04</td>
<td>99.4</td>
<td>0.5</td>
<td>7.96±0.06</td>
</tr>
<tr>
<td>Soil (2g/100 mL)</td>
<td>---</td>
<td>183.5±0.7</td>
<td>0.38</td>
<td>183.2±0.8</td>
<td>---</td>
</tr>
<tr>
<td></td>
<td>30</td>
<td>213.4±0.6</td>
<td>99.9</td>
<td>0.28</td>
<td>213.0±0.6</td>
</tr>
</tbody>
</table>

² Mean ± standard deviation (n = 5).
³ Tabulated F-value for (4,4) degrees of freedom at P (0.95) is 6.39.
⁴ Tabulated t-value for 8 degrees of freedom at P (0.95) is 2.306.
5B.4. CONCLUSIONS

The sensitivity, simplicity, temperature independence, stability of the formed azo dye (4 h), wide range of chromium determination and high tolerance towards a large number of foreign ions are the advantages of the proposed method. Since no extraction step is required, the use of organic solvents, which are generally toxic pollutants, is avoided. The method is validated by the parallel determinations with standard diphenylcarbazide method. The statistical analysis showed that, there is no significant difference between the proposed and the standard methods. The method developed can serve as an alternative method for determining chromium in various samples.

The proposed methods are compared with many other reported methods (Table 5.10). It shows that the developed methods are more sensitive and simple compare to most of the mentioned methods in the table.
**Table 5.10.** Comparison of the proposed method with some other spectrophotometric methods

<table>
<thead>
<tr>
<th>Reagents</th>
<th>Ref.</th>
<th>( \lambda_{\text{max}} ) (nm)</th>
<th>( \varepsilon ) (L mol(^{-1}) cm(^{-1}))</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>Prochlorperazine dimaleate (PCPM)</td>
<td></td>
<td>535</td>
<td>(2.09 \times 10^4)</td>
<td>Simple, sensitive, no extraction or heating is required and less interference.</td>
</tr>
<tr>
<td>Citrazinic acid (CZA)</td>
<td>Proposed methods</td>
<td>515</td>
<td>(2.1 \times 10^4)</td>
<td></td>
</tr>
<tr>
<td>4-(2-Thiazolylazo)- resorcinol</td>
<td>[22]</td>
<td>545</td>
<td>(2.72 \times 10^4)</td>
<td>Surfactant (CTAB) was used, Fe(III) strongly interfere.</td>
</tr>
<tr>
<td>1,4,8,11-tetraazacyclo tetradecane</td>
<td>[24]</td>
<td>379</td>
<td>(1.5 \times 10^4)</td>
<td>Less sensitive, requires a reaction time of 10 min.</td>
</tr>
<tr>
<td>Chromotropic acid</td>
<td>[29]</td>
<td>355</td>
<td>(1.76 \times 10^4)</td>
<td>Less sensitive, oxidizing agents interfere.</td>
</tr>
<tr>
<td>Leuco xylene cyanol FF</td>
<td>[30]</td>
<td>615</td>
<td>(8.23 \times 10^4)</td>
<td>Sensitive, required heating at 90 °C for 15 min, Fe(III), Ce(IV) and V(V) interfere severely.</td>
</tr>
<tr>
<td>Magneson I</td>
<td>[31]</td>
<td>420</td>
<td>(1.02 \times 10^4)</td>
<td>Less sensitive, oxidizing agents interfere.</td>
</tr>
<tr>
<td>Perphenazine</td>
<td>[34]</td>
<td>526</td>
<td>(1.87 \times 10^7)</td>
<td>Less sensitive, susceptible to other redox species.</td>
</tr>
<tr>
<td>Chlorpromazine-HCl</td>
<td>[35]</td>
<td>526</td>
<td>(3.28 \times 10^4)</td>
<td>Less selective, highly susceptible to other redox species.</td>
</tr>
<tr>
<td>Chlorophosphonazo I</td>
<td>[38]</td>
<td>580</td>
<td>(1.33 \times 10^4)</td>
<td>Less sensitive, time consuming.</td>
</tr>
<tr>
<td>Pentamethylene bis-(triphenyl phosphonium) bromide</td>
<td>[53]</td>
<td>365</td>
<td>(1.38 \times 10^3)</td>
<td>Less sensitive, extractive, Mn(VII), Mo(VI) and W(VI) interfere.</td>
</tr>
<tr>
<td>N-methylpiperazine-4-carbodiithioate</td>
<td>[54]</td>
<td>410</td>
<td>(5.24 \times 10^7)</td>
<td>Less sensitive, extractive, required heating at 60 °C for 5 min, EDTA, Fe(III), Co(II) and Cu(II) interfere</td>
</tr>
<tr>
<td>Hydroxyamidine</td>
<td>[55]</td>
<td>395</td>
<td>(1.45 \times 10^4)</td>
<td>Less sensitive, extractive, several metal ions interfere.</td>
</tr>
<tr>
<td>(p)-methylisonitroso acetophenone</td>
<td>[63]</td>
<td>430</td>
<td>(8.056 \times 10^3)</td>
<td>Less sensitive, extractive, several metal ions interfere.</td>
</tr>
<tr>
<td>Phenanthraquinone</td>
<td>[64]</td>
<td>470</td>
<td>(6.65 \times 10^3)</td>
<td>Less sensitive, extractive.</td>
</tr>
<tr>
<td>Rhodamine 6G</td>
<td>[70]</td>
<td>535</td>
<td>(2.0 \times 10^4)</td>
<td>Extractive (toluene), Fe(III), Fe(II) and Mn(VII) interfere.</td>
</tr>
<tr>
<td>N-methyliniline carbodiithioate</td>
<td>[71]</td>
<td>340</td>
<td>(8.2 \times 10^4)</td>
<td>Less sensitive, extractive.</td>
</tr>
</tbody>
</table>
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