A New Method for the Direct Determination of Dissolved Fe(III) Concentration in Acid Mine Waters

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ABSTRACT

A new method for direct determination of dissolved Fe(III) in acid mine water has been developed. In most present methods, Fe(III) is determined by computing the difference between total dissolved Fe and dissolved Fe(II). For acid mine waters, frequently Fe(II) >> Fe(III); thus, accuracy and precision are considerably improved by determining Fe(III) concentration directly. The new method utilizes two selective ligands to stabilize Fe(III) and Fe(II), thereby preventing changes in Fe reduction-oxidation distribution. Complexed Fe(II) is cleanly removed using a silica-based, reversed-phase adsorbent, yielding excellent isolation of the Fe(III) complex. Iron(III) concentration is measured colorimetrically or by graphite furnace atomic absorption spectrometry (GFAAS). The colorimetric method requires inexpensive commercial reagents and simple procedures that can be used in the field. The method detection limit is 0.002 mg/L (40 nM) using GFAAS, and 0.02 mg/L (0.4 µM) by colorimetry.

INTRODUCTION

Accurate and precise measurements of Fe reduction-oxidation (redox) species are particularly important in the study of acid mine waters because Fe is a major component in such waters. Charge balances calculated for acid mine waters depend strongly on the Fe(II)/Fe(III) ratio. Aqueous speciation is sensitive to the absolute concentrations of Fe(II) and Fe(III) as well as the Fe(II)/Fe(III) ratio. Iron(III) precipitates readily, forming hydrous ferric oxide, which adsorbs trace metals. Thus, Fe controls the mobility and toxicity of other metals. A method for determining Fe(III) concentration is needed to accurately predict the fate and mobility of metals in high-Fe aquatic environments.

Dissolved Fe(II) concentrations in waters are commonly determined by a colorimetric technique using a ferroin complexing reagent; it is also preferable to determine total dissolved Fe (Fe_T) concentrations by colorimetry. In a study comparing analytical methods for the determination of major and trace constituents in acid mine waters, Ball and Nordstrom (1993) found that colorimetric determination of Fe_T, using FerroZine®,[1] as the complexing agent, was more reliable, precise, and sensitive than inductively-coupled plasma or direct-current plasma spectrometry.

Most methods for determining Fe(III) concentration are based on colorimetric determination of the Fe(II) concentration, followed by a separate determination of the Fe_T concentration after reduction of Fe(III) (Greenberg and others, 1992). The difference between the concentrations of Fe_T and Fe(II) is taken as the Fe(III) concentration. One major problem with this approach is that both accuracy and precision of the Fe(III) concentration are compromised as the proportion of Fe(II) increases, with determinations often overestimating actual Fe(III) concentrations. Acid mine waters usually contain much more Fe(II) than Fe(III), making the difference between Fe_T

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[1] Registered trademark of the Hach Company, Loveland, CO. The use of trade, brand, or product names is for identification purposes only and does not constitute endorsement by the U.S. Geological Survey

and Fe(II) comparable to the error of the determination.

Prompt on-site determination of the Fe species may be important because the relative concentrations of the Fe redox states may change in the presence of oxygen, light, and biocatalysts (Nordstrom and Alpers, 1999). Portable UV-Visible spectrophotometers allow colorimetric Fe measurements to be taken immediately after sample collection.

While determination by difference of one Fe valence state is a simple matter when $0.2 \leq Fe(II)/Fe(III) \leq 5$, it becomes far more difficult at very large or very small ratios. In such instances, reagents are needed that are specific for the respective oxidation states. The proposed method uses selective complexing agents for both Fe(II) and Fe(III). The new method corrects for the shortcomings of many reported methods and current practices, and provides accurate and precise measurements of Fe(III) in the presence of other metals including Fe(II). Desirable features of the method include reliability, portability, and low costs.

Acetohydroxamic acid was selected as the Fe(III) chelator because it has a high selectivity for Fe(III) (Raymond and others, 1984; Hider and Hall, 1991; Purohit and others, 1994), it is commercially available, and it is inexpensive. Derivatives of this compound could be used to further enhance the molar absorptivity and sensitivity of the method.

EXPERIMENTAL

Sample Collection and Preservation

Samples collected in the field are immediately filtered through a 0.1 µm tortuous-path membrane, acidified to pH of about 1 with hydrochloric acid (2 mL 6 M HCl per 250 mL sample), and stored in acid-washed opaque plastic bottles at 4°C. Samples collected and preserved in this manner may be stored for up to 6 months without significant changes in the Fe redox distribution because microbial catalysts are removed, the pH is low enough to keep metals solubilized, and the iron oxidation rate is negligible.

Apparatus

A diode-array spectrophotometer (HP8452A) with 5cm cells, Zeeman Atomic Absorption Spectrometer (PE 4110ZL), Alltech maxi-clean C18 cartridges containing 900 mg of absorbent, and plastic syringes (10-mL and 30-mL) were used.

Reagents

Double-distilled water; 1.0 M acetohydroxamic acid ($C_2H_4NO_2$), Aldrich, 98%; 4.9×10^{-3} M FerroZine iron reagent ($C_{20}H_{13}N_4S_2O_6Na\cdot H_2O$), Hach, 95.2% pure; 6 M redistilled hydrochloric acid (HCl); methanol (CH_3OH), 99.9%, A.C.S. HPLC grade; hydroxylamine hydrochloride solution ($NH_2OHAHCL$), 10% w/v; and ammonium acetate buffer solution (CH_3COONH_4), pH 7-7.5. The buffer was prepared by diluting 467 mL reagent grade (28-30%) ammonium hydroxide (NH_4OH) and 230 mL ultrapure glacial acetic acid (CH_3COOH) to 1 L. Iron(III) standard stock solution: 10.0 mg/L as ferric sulfate hexahydrate ($Fe_2(SO_4)_3\cdot 6H_2O$) in double distilled water containing 1% (v/v) 6 M redistilled HCl. The Fe in the standard was greater than 99.0% Fe(III), as determined by the FerroZine method. Iron(II) standard stock solution: 100 mg/L as ferrous ammonium sulfate hexahydrate ($Fe(NH_4)_2(SO_4)_2\cdot 6H_2O$) in double distilled water containing 1% (v/v) 6 M redistilled HCl. The Fe in the standard was greater than 99.0% Fe(II), as determined by the FerroZine method.

The following single-element standards were used for interference studies: Cu(II), 1,000 ± 3 mg/L in 2% HNO_3; Ca(II), 10,000 ± 5 mg/L in 5% HNO_3; Cr(III), 1,000 ± 3 mg/L in 5% HCl and 1% HNO_3; Al(III), 1,000 ± 5 mg/L in 5% HCl and 1% HNO_3; Co(II), 1,000 ± 3 mg/L in 2% HNO_3; Ni(II), 1,000 ± 5 mg/L in 5% HNO_3; Pb(II), 1,000 ± 3 mg/L in 2% HNO_3; Zn(II), 1,000 ± 5 mg/L in 5% HNO_3; and Cd(II), 1,000 ± 3 mg/L in 2% HNO_3.

Procedure

1. Determine dissolved Fe(II) and Fe_T using the FerroZine method as follows:

a. Pipette an adequate volume of acidified sample (up to 20 mL maximum) which will yield 0.004-1.6 mg/L Fe when diluted to volume, into a 25-mL volumetric flask.

b. Prepare appropriate blanks and standards, in 25-mL volumetric flasks.

c. If solution is a standard or blank, add 0.25 mL hydroxylamine hydrochloride solution (reduces Fe(III) to Fe(II)).

d. Add 0.5 mL FerroZine reagent and mix (the FerroZine forms a complex with Fe(II)).

e. Add 1.25 mL buffer solution (buffers the pH near neutrality), rinse down neck of flask, shake well, and allow at least 5 min for full color development.

f. If the magenta color of the FerroZine complex is masked or discolored by a brick or rust red color due to the presence of high Fe(III) concentrations, add 1.50 mL 6 M HCl, after color development. If Fe(III) interference is suspected but not noticeable to the eye, check duplicates, one with acid against one without: absorbances should be nearly identical if no interference is present.

g. Dilute to the mark and shake well.

h. Measure absorbance at 562 nm. Solutions must be measured within 1-2 hrs.

i. For Fe_T, use the same procedure with the addition of step 1.c. to all samples.

2. Transfer a maximum of 18.25 mL of sample containing no more than 14 mg/L Fe(II) and no more than 1.2 mg/L Fe(III) when diluted to volume to a 25-mL volumetric flask.

3. Add the following reagents in the order listed:

a. 5.0 mL FerroZine solution.

b. 0.5 mL acetohydroxamic acid (the acetohydroxamic acid will form a complex with Fe(III)).

c. 1.25 mL ammonium acetate buffer (buffers the pH near neutrality).

4. Dilute to volume with double-distilled water.

5. Remove the Fe(II)-FerroZine complex from the sample as follows:

a. Attach a new or regenerated C_{18} cartridge to a 10-mL plastic syringe.

b. Condition C_{18} cartridge by passing through it 5 mL methanol followed by 5 mL double-distilled water. Do not allow cartridge to dry before sample processing.

c. Remove C_{18} cartridge from syringe and attach it to a new 30-mL syringe.

d. Remove plunger, transfer sample into the syringe, then force sample through cartridge at a fast, steady drip (2-3 drops per second). Discard first 5 mL of filtrate and collect remaining 20 mL for Fe(III) determination.

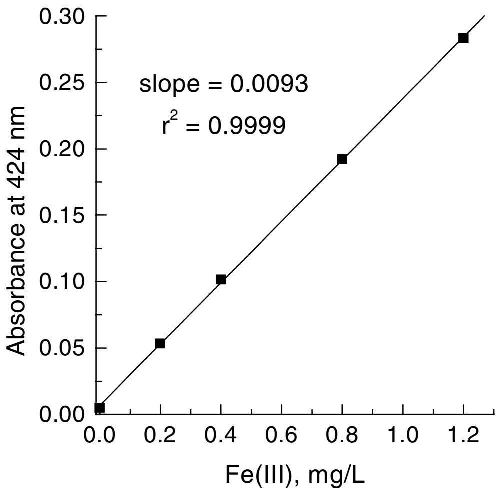
6. Determine the Fe(III) concentration colorimetrically (within 5 hours) or by Zeeman-corrected graphite furnace atomic absorption spectrometry (GFAAS).

7. Regenerate C_{18} cartridge by passing 10 mL methanol through it.

RESULTS AND DISCUSSION

A typical calibration curve for Fe(III) by colorimetric analysis is shown in figure 1. The visible absorption spectrum of the Fe(III)-acetohydroxamic acid complex exhibits a single peak with maximum absorbance at 424 nm. At this wavelength, the molar absorptivity is 2,583 $cm^{-1} mol^{-1}$ and Beer's law is obeyed to approximately 16 mg/L. The Fe(III)-acetohydroxamic acid complex is stable for up to 5 hours. The redox stability of the combined Fe(II) and Fe(III) complexes was tested by time series determinations, and no change in Fe(II)/Fe(III) ratio was found for up to 3 hours.

Figure 1. Standard curve for colorimetric



determinations

Accuracy of Direct Fe(III) Measurements in Acid Mine Waters

The accuracy of the new method was estimated by performing spike recoveries. Two or three additions of Fe(III) standard solution were added to six different samples and treated with the described procedure. The samples were collected from three different acid mine effluents: Summitville Mine, Rio Grande County, CO; Paradise Portal in the Upper Animas Mine Drainage, San Juan County, CO; and Iron Mountain Mine, Shasta County, CA. GFAAS determinations were performed for samples found to contain less than 0.02 mg/L Fe(III) by colorimetric analysis. Recoveries were 93-101% for colorimetric determinations and 89-91% for GFAAS (fig. 2, table 1).

Table 1. Iron(III) spike recoveries using the acetohydroxamic acid method.

Sample ID	Fe(III) added mg/L	Recovered mg/L	%	Analysis
Alamosa River below Terrace Reservoir	0.010	0.009	91	GFAAS
	0.020	0.018	89	GFAAS
Spring Creek at weir	0.100	0.100	100	Color
	0.200	0.203	101	Color
	0.400	0.403	101	Color
Chandler Adit of Summitville Mine	0.100	0.097	97	Color
	0.200	0.197	98	Color
	0.400	0.400	100	Color
Paradise Portal	0.100	0.093	93	Color
	0.200	0.198	99	Color
	0.400	0.384	96	Color
Keswick Reservoir Pore Water	0.100	0.097	97	Color
	0.200	0.198	99	Color
	0.400	0.396	99	Color

Analytical Considerations

The C_{18} cartridges retain some Fe(II)-FerroZine complex. Thus, for best accuracy and precision at Fe(III) concentrations below about 0.2 mg/L, it is critical to pre-clean C_{18} cartridges by processing a blank solution through them. The C_{18} cartridges can be regenerated virtually indefinitely when used to determine Fe(III) concentrations higher than 0.2 mg/L. The pH of the final solution should be between 4 and 7. The

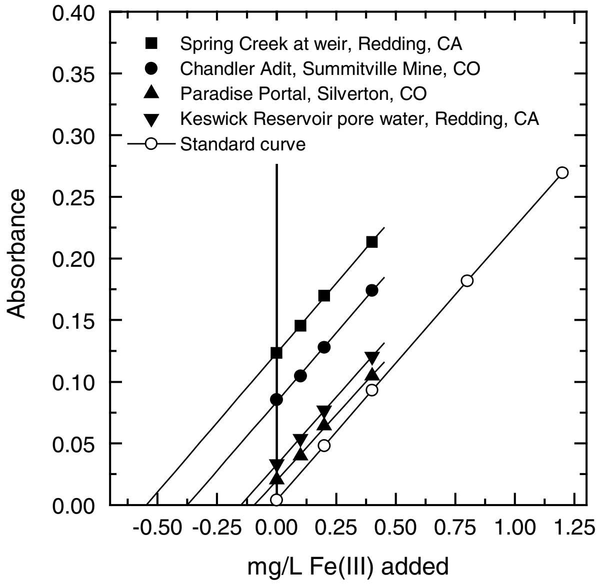


Figure 2. Method of standard additions for Fe(III) using the acetohydroxamic acid method.

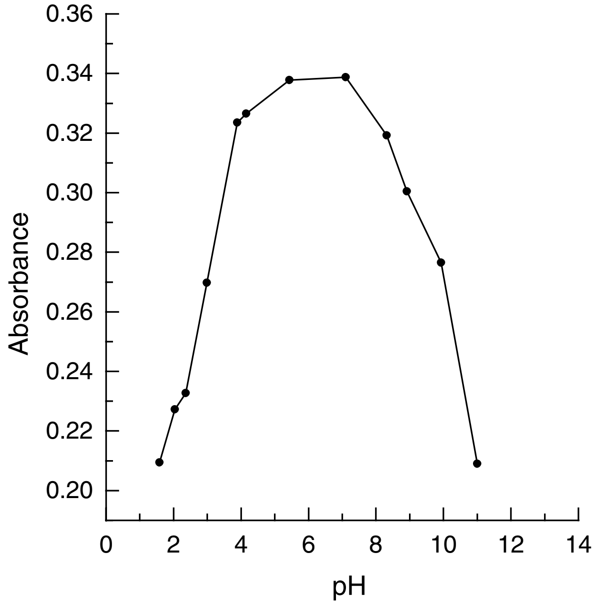


Figure 3. Effect of pH on the formation of Fe(III)-acetohydroxamic acid complex.

yellow Fe(III)-acetohydroxamic acid complex (fig. 3) and the Fe(II)-FerroZine (Stookey, 1970) complex will form completely in aqueous solution in this pH range.

The detection limit by colorimetric analysis using a 5-cm cell is 0.02 mg/L Fe(III). Standards for colorimetric determinations were prepared in the same way as samples and contained 0, 0.2, 0.4, 0.8, and 1.2 mg/L Fe(III) when diluted to volume. The detection limit by Zeeman-corrected GFAAS is 0.002 mg/L Fe(III). The less-sensitive colorimetric analysis is preferred when Fe(III) concentrations are above its detection limit and results are needed in real time, such as during field studies.

The sample should be filtered through a 0.1 µm or smaller pore-sized membrane. In a study of filter pore-size effects on the analysis of dissolved Fe, Al, Mn, and Ti in natural water, errors of an order of magnitude or more in the measurement of the dissolved metals were found when a 0.45 µm filter was used (Kennedy and others, 1974). This was caused by fine-grained particulate material passing through the membrane. Compared with a 0.45 µm membrane, the 0.1 µm membrane reduces the passage of particulate materials without significantly increasing filtration time. More recently, investigators have demonstrated that iron colloids also can pass through a 0.1 µm membrane (Kimball and others, 1995).

Potential Interferences

The FerroZine and ammonium acetate reagents did not interfere with the Fe(III)-acetohydroxamic acid complex. Acid mine water may contain at least 31 major chemical species with concentrations that vary up to several orders of magnitude. Many of those constituents may cause interferences in the colorimetric determination of Fe(III). Ten metals found in a typical acid mine effluent that may cause interferences have been tested. Chromium(III) was selected because of its chemical similarity to Fe(III). The remaining metals were selected based on their reported binding constants with acetohydroxamic acid (Martell and Smith, 1976).

The level of interference by each metal with the quantitation of Fe(III) was determined by using the new method to measure the change in apparent Fe(III) content in the presence of each metal. Individual solutions containing zero or 0.4 mg/L Fe(III) were analyzed in triplicate with up to 40 mg/L of potential interferent, except for Ca(II) (400 mg/L), added. The change in apparent Fe(III) content in the presence of 40 mg/L Al(III), Pb(II), Zn(II), Ni(II), and Cd(II), and 400 mg/L Ca(II) was less than ±0.008 mg/L. This change is considered insignificant because it is well below the minimum difference in Fe(III) content measurable by the new method.

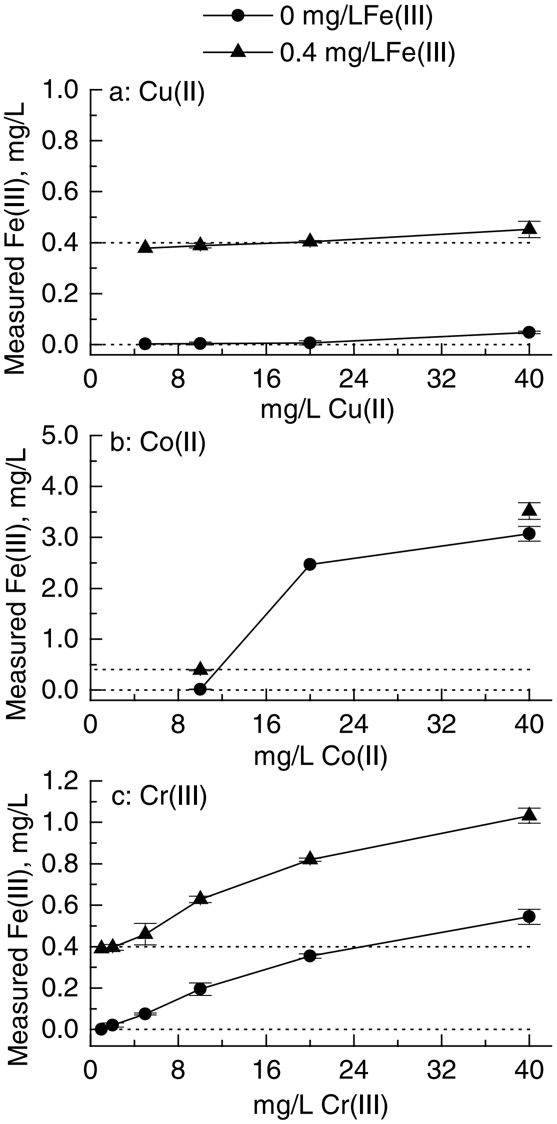


Figure 4. Apparent Fe(III) concentration in the presence of added Cu(II), Co(II) and Cr(III). [error bars represent ± 1s for triplicate determinations]

The apparent Fe(III) content increases by 0.052 mg/L in the presence of 40 mg/L Cu(II). The tested Cu(II)/Fe(III) molar ratio was 88, whereas the ratio found in acid mine samples is typically less than 1. Of 25 samples taken from the Leviathan Mine drainage basin, located in California and Nevada, the average Cu(II)/Fe(III) ratio was 0.18 with 2 samples having a ratio slightly greater than 1 (Ball and Nordstrom, 1989). In the presence of 20 mg/L Cu(II), the change in apparent Fe(III) content was less than 0.008 mg/L (fig. 4a).

Co(II) forms a colored complex with FerroZine (Dawson and Lyle, 1990) which is not completely retained by the C_{18} cartridge at Co(II) > 10 mg/L and absorbs intensely at wavelengths less than 350 nm. The tail of the absorption band interferes with the 424-nm line. The highest Co(II)/Fe(III) molar ratio in the 25 Leviathan Mine drainage basin samples discussed above is 0.8. In the presence of 10 mg/L Co(II) (Co(II)/Fe(III) molar ratio of 24) the Co(II)-FerroZine complex is completely retained by the C_{18} cartridge. The change in apparent Fe(III) content was about +0.016 mg/L for a blank and about –0.008 mg/L in a 0.4 mg/L standard (fig. 4b). These deviations are not significant relative to the measured precision.

The apparent Fe(III) content increased by about 0.6 mg/L in the presence of 40 mg/L Cr(III). In the presence of 2 mg/L Cr(III) (Cr(III)/Fe(III) molar ratio of about 5), no change in the apparent content of Fe(III) in test solutions could be detected (fig. 4c). In most acid mine effluent samples the Cr(total)/Fe(III) molar ratio is usually well below 0.5. Chromium, Co, and Cu are expected to interfere only under unusual conditions.

Acetohydroxamic acid can oxidize Fe(II) to Fe(III), causing overestimation of the Fe(III) concentration. To control this source of error, it is necessary to add the FerroZine reagent before the acetohydroxamic acid so that the distribution of Fe redox species is stabilized. Separation of the two Fe complexes is necessary to prevent the strongly absorbing Fe(II)-FerroZine complex from interfering with the colorimetric determination of Fe(III). The Fe(II)-FerroZine complex exhibits a single peak with a maximum absorbance at 562 nm (Stookey, 1970). The C_{18} cartridge retains the more hydrophobic Fe(II)-FerroZine complex while allowing the more hydrophilic Fe(III)-acetohydroxamic acid complex to pass into the effluent. The 562-nm absorbance remained at the baseline for synthetic Fe samples containing 0, 0.2, and 0.8 mg/L Fe(III) and 0 to 20 mg/L Fe(II), illustrating the completeness of the separation.

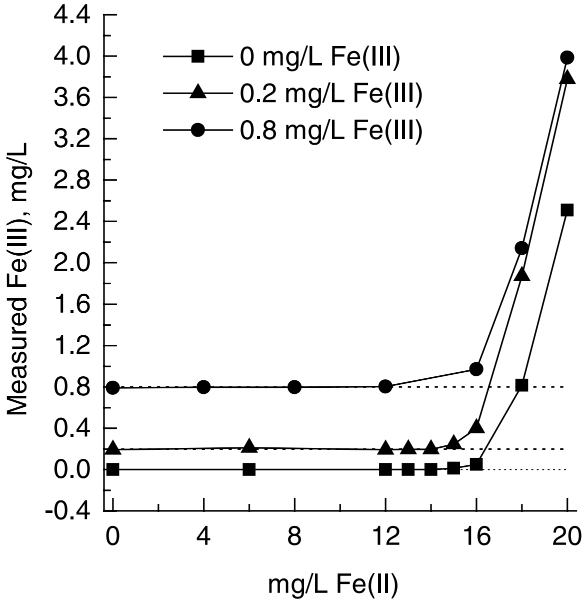


Figure 5. Iron(II) interferences and tolerance limits for direct determination of Fe(III) using the acetohydroxamic acid method.

Table 2. Lower limits of the acetohydroxamic acid method in the presence of Fe(II).

	Lowest Fe(III) concentration (mg/L) that can be detected by:	
Fe(II), mg/L	**Colorimetric**	**GFAAS**
10,000	11.3	1.13
2,000	2.27	0.227
500	0.57	0.057
100	0.11	0.011
<20	0.02	0.002

Under the conditions stated in the procedure, Fe(II) concentrations less than14 mg/L do not interfere with the Fe(III) determination (fig. 5). The stated quantity of FerroZine has the capacity to complex a maximum of 350 µg Fe(II). More FerroZine could be used, but the capacity of the C_{18} cartridge would be exceeded. Excess Fe(II) is oxidized and forms Fe(III)-acetohydroxamic acid. Iron(III) was separated (97-103% recovery) from synthetic iron samples containing Fe(II)/Fe(III) ratios from 0 to greater than 500. Using the stated procedure, Fe(III) can be measured colorimetrically in solution with Fe(II)/Fe(III) ratios up to 880, and by GFAAS in solutions with ratios up to 8800 (table 2).

Comparing the FerroZine Method with the New Method

The performance of the new method was compared with that of the FerroZine method for Fe(III) determinations. Four representative samples were selected for the comparison. Values of percent difference (Δ%), equation (1):

$$\Delta\% = 100 \times \frac{C_A - C_B}{(C_A + C_B) \div 2} \qquad (1)$$

were used for the comparison, where C_A is concentration determined by the FerroZine method and C_B is concentration determined by the new method with either colorimetric or GFAAS determinations. The maximum value of this function is ± 200%. A Δ% value of 0 denotes a perfect match of the analytical values, while a value approaching ± 200 means there is no similarity between values (Ball and Nordstrom, 1993). A positive Δ% value indicates that Fe(III) concentration determined by FerroZine is greater than the concentration measured by the new method. To compare the analytical results of direct Fe(III) determinations with the Fe(III) results obtained by difference using the FerroZine method, values of precision and the Δ% function were calculated. Iron(III) concentrations obtained by difference using the FerroZine method are typically greater than those obtained using the new method. The value of the Δ% function was +32% in one case. A graphical representation of results for selected samples illustrating the dramatic improvement in precision using the new method is shown in figure 6.

The power of the new method lies in its capability to determine Fe(III) concentration in samples containing very high Fe(II)/Fe(III) ratios. As the Fe(II)/Fe(III) ratio increases, the relative standard deviation of Fe(III) concentrations obtained by difference generally increases. Relative standard deviations for Fe(III) determinations using the FerroZine method for 50 different samples collected from three different acid mine effluents including Summitville Mine, Rio Grande County, CO; Paradise Portal in the Upper Animas Mine Drainage, San Juan County, CO; and Iron Mountain Mine, Shasta County, CA increased to over 50 percent in samples with Fe(II)/(III) ratios of 30 or higher. This uncertainty is often too large for Fe(III) concentrations determined by difference to be meaningful. The relative standard deviations for Fe(III) concentrations determined directly using the new method are generally less than 5 percent.

A more detailed description of the method can be found in To and others (1999).

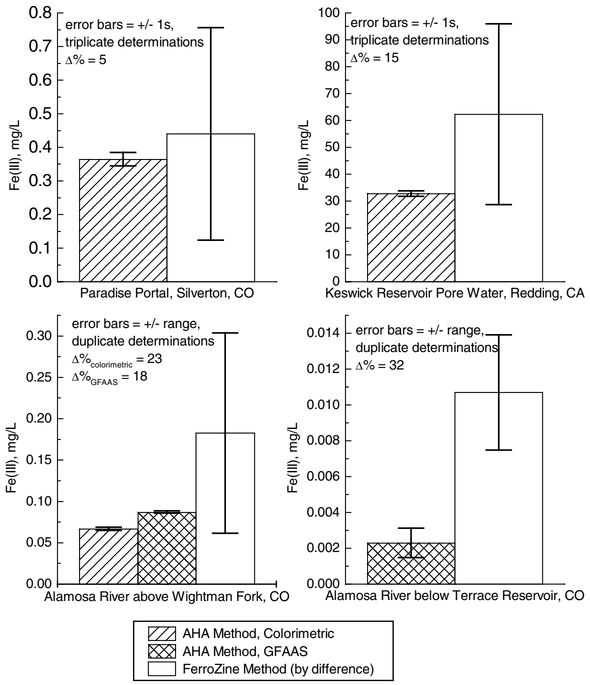


Figure 6. Comparison of Fe(III) concentrations determined directly using the acetohydroxamic acid (AHA) method with Fe(III) concentrations determined by difference using the FerroZine method.

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