# Methods for handling redox-sensitive smectite suspensions

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## ABSTRACT

For handling and storage of redox-sensitive smectites, a controlled atmosphere liquid exchange (CALE) apparatus and glove box were used. For this purpose a fine fraction of $Na^+$-SWa-1 ferruginous smectite was used in this work. Reduction of structural Fe was conducted with sodium dithionite and buffered by sodium citrate-sodium bicarbonate (C-B) solution at 70 °C for 4 h. Changes in color from yellow-brown of unreacted through green-yellow (partially reduced) to dark grey (totally reduced) were observed. A series of prepared samples were washed with de-oxygenated NaCl solutions without contact with oxygen, which could lead to re-oxidation. Another series of samples were washed with NaCl solutions with higher oxygen content. Higher efficiency and suitability of the CALE apparatus for $O_2$ sensitive clay redox systems was demonstrated. It was more effective in reaction products removal and maintaining of higher Fe

reduction. Analysis of solid phases by 1, 10-phenanthroline method provided a high conversion level (91.129 ± 0.477 % of Total Fe %).

**KEY WORDS:** nontronite**,** iron, reduction, re-oxidation, controlled atmosphere liquid exchange apparatus, glow box, centrifuge,

## INTRODUCTION

Accurate research in all fields of investigation requires precise tools and techniques to eliminate errors as much as possible. This is especially true for materials (solid, liquid, and/or gaseous) that can be modified by exposure to the natural environment ($O_2$, humidity, temperature, pressure, UV radiation, etc.) during their sampling, handling, analysis, and storage. Redox-sensitive clay minerals certainly fit within this category.

In general, the reduction of Fe(III) to Fe(II) in Fe-rich smectites is now well known (Stucki, 2006, 2013) and has been the aim of numerous investigations over about four decades (Rozenson and Heller-Kallai, 1976; Stucki and Roth, 1977; Russell *et al.*, 1979; Stucki *et al.*, 1984a; Wu et al., 1989; Komadel *et al.*, 1990, 1995, 2006; Stucki and Tessier, 1991; Manceau *et al.*, 2000; Stucki *et al.*, 2002; Hofstetter *et al.*, 2003; Merola *et al.*, 2007; Schaefer *et al.*, 2011; (Hofstetter *et al.*, 2008; Neumann *et al.*, 2008; Dong *et al.*, 2009; Neumann *et al.*, 2009; Gorski *et al.*, 2011; Neumann *et al.*, 2011; Alexandrov *et al.*, 2013), and others). This phenomenon has also been investigated and described as it relates to both biological (Stucki *et al.*, 1987; Kostka *et al.*, 1999; Dong *et al.*, 2003, 2009; Li *et al.*, 2004; Jaisi *et al.*, 2005, 2007; O'Reilly *et al.*, 2005, 2006; Kukkadapu *et al.*, 2006; Lee *et al.*, 2006; Zhang *et al.*, 2007; Ribeiro *et al.*, 2009; Pentráková et al., 2013) and environmental (Peretyazhko *et al.*, 2008; Jaisi *et al.*, 2009; Neumann

*et al.*, 2009; Zhang *et al.*, 2009; Cervini-Silva *et al.*, 2010; Bishop *et al.*, 2011; Southam, 2012; Zhuang *et al.*, 2012) processes.

An understanding of these phenomena over the past decades came about because of the development of methods and apparatus which preserve the oxidation state of the structural Fe during preparation and analysis of the samples. The need for these methods was recognized by Stucki (1975) while attempting to confirm the oxidation-reduction mechanism for structural Fe in smectites proposed by Roth *et al.* (1969) and Roth and Tullock (1972), which was based on the belief that no change in layer charge occurred during reduction of the structural Fe. By protecting the sample from atmospheric $O_2$ during measurements of cation exchange capacity, however, Stucki and Roth (1977) showed a definitive increase in layer charge with increasing structural Fe(II), but that the increase was not a linear function of the extent of reduction. The methods and apparatus used in those studies were described briefly by Stucki et al. (1984a), but a complete and detailed description has never been given, especially one that includes how they have evolved and been improved through the years. This is the purpose of the present publication.

For correct characterization of such materials, they must be sampled, handled, and analyzed under conditions which prevent or minimize re-oxidation. Described below are laboratory methods and apparatus that accomplish these aims, and include methods for chemical reduction, removal of excess salts, addition of $O_2$-free reactants, drying, sample transfer, storage, and analysis by chemical and spectroscopic methods.

## MATERIALS AND METHODS

### Materials

Redox-protective methods were illustrated and tested using ferruginous smectite (sample SWa-1 from the Source Clays Repository of The Clay Minerals Society) as the example clay mineral submitted to a chemical reduction process which included various steps of washing and freeze drying. The sample was fractionated (<2 μm), $Na^+$-saturated, and freeze dried before use and labeled Na-SWa-1 (Stage 0, Figure 1). All chemicals used were analytical-reagent grade, except sodium dithionite was technical grade. The supplier of all chemicals was Thermo-Fisher Scientific (Pittsburgh, Pennsylvania). Citrate-bicarbonate (C-B) buffer solution was prepared by combining 24 parts of 1.0 M sodium bicarbonate and 1 part of 0.9 M sodium citrate. All water used was purified to a resistivity of approximately 18 MOhm-cm and is referred to hereafter as DI water.

## Methods

### *$O_2$-free gas*

Oxygen-free gas was obtained by passing standard Ar gas through a hot (130 °C) $O_2$ trap (Chromatography Research Supplies (CRS), Louisville, Kentucky, ¼-inch Model 1000 [or older bugle-shaped version] high-capacity $O_2$ trap with 500 $cm^3$ of Cu/Zn/Al catalyst) wrapped with heating tape and covered with aluminum foil. Valves were installed at both ends of the trap. The purge gas could be $N_2$ (less expensive than Ar) if permitted by the objectives of the study. In the authors' laboratory, Ar was selected over $N_2$ because some target analyses were for species of N. The CRS Model 1000 $O_2$ trap has a capacity of 2.5 L of $O_2$ at room temperature and xx L at 130 °C.

Once spent, the catalyst in the trap required regeneration. This was accomplished by pre-heating it to 130 °C then purging with $N_2$ for 15 min at a flow rate of 50 $cm^3$/min. The purge gas was then changed to a mixture of Ar + 10% $H_2$, the temperature raised to 300 °C, and the flow

rate increased to 350 $cm^3$/min. The trap was then held under these conditions for 4 h. The effluent gas was vented into a hood. Water formed as the $H_2$ reacted with adsorbed $O_2$, and the inert gas swept the water vapor from the trap. When regeneration was complete, no more water vapor could be detected in the gas stream. The outlet valve to the trap was then closed and the line at that end disconnected. The power was turned off to allow the trap to cool under regenerating gas pressure. When the trap reached room temperature, the input valve was closed and the trap was disconnected from the regeneration gas input line and reinstalled at the site of application.

To reinstall the $O_2$ trap at the application site without exposing the catalyst to the atmosphere or contaminating the inert-gas supply with atmospheric $O_2$, the following procedure was used. First, all valves in the inert-gas supply line downstream from the main gas valve on the cylinder and between it and the $O_2$ trap were opened to the atmosphere and the line was disconnected at the inlet end of the trap. Second, the main gas valve on the cylinder was opened to sweep all atmospheric gases away from the main cylinder supply. Third, the flowing stream of the inert input gas was flooded over the input connector to the trap, which was then connected and tightened while the gas was flowing. Fourth, the input valve to the trap was opened. Fifth, the heating tape around the trap body was energized to raise the catalyst temperature to 130 °C. Sixth, the output valve was not opened until needed to purge the target apparatus. This sequence is designed to minimize the contamination of the inert gas supply and to extend the life of the catalyst in the trap.

***Apparatus for handling Fe-reduced clay***

Fundamental to the investigation of redox-modified smectites is an inert-atmosphere reaction tube (IRT), which consisted of a centrifuge tube and septum-sealing cap (Figure 1A) in

which the clay mineral sample was dispersed, reduced, washed, and reacted with desired chemical solutions. It was designed so that the dispersion could be accessed using septum-penetration needles (6 inch, 22 gauge, deflected point), which enabled solution manipulations without exposure to the atmosphere. For this purpose, a 50-mL polycarbonate, round-bottom, Oak Ridge type centrifuge tube was chosen (Figure 1A, 17), such as Thermo-Fisher catalog number 3118-0050. It has the advantages of chemical resistance, transparency, and reasonable volume. A septum-sealing cap to fit this centrifuge tube was designed with three parts: (1) a rigid teflon disc with o-ring (such as Thermo-Fisher catalog number DS3131-0024) and a 0.25-inch diameter center hole (hand drilled in the laboratory) (Figure 1A, 15), which was placed on the opening of the centrifuge tube such that the o-ring sealed against the lip of the tube when tightened; (2) a septum disc (Figure 1A, 14) cut from a Pursep T® septum sheet (catalog number 230696, Chromatography Research Supplies, Louisville, Kentucky, USA, [describe septum materials here]), using a 1-inch diameter, hollow-cylinder cutter (such as a cork borer), which was then placed across the opening of the centrifuge tube, supported by the rigid teflon disc; and (3) an Al cap (Figure 1A, 13), also drilled with a 0.25-inch center hole, to compress and seal the septum and septum support onto the centrifuge tube. All pieces were designed to fit inside a Dupont/Sorvall model SS-34 centrifuge rotor or equivalent.

The septum-penetration needles used to gain access into the IRT were attached to the sampling port of a controlled-atmosphere liquid exchanger (CALE) (Figure 2). The sampling port was connected to various condenser flasks containing degassed solutions, which could be individually selected, through a carefully orchestrated network of valves and tubing (Figure 2). The CALE used in the authors' laboratory contained four flasks in which four different solutions could be deoxygenated before being selected for solution transfer into the IRT.

Each condenser flask in the CALE was a modified 2-L flat bottom flask (Figure 1B, 2) to which four equally spaced vertical ports were added using ACE-Threds glass risers to accept 0.25-inch glass tubing. Sealed access to the inside of the flask was gained through these ports for a gas dispersion tube (Figure 1B, 8), which was slightly curved away from the flask wall; a solution withdrawal tube (Figure 1B, 7), which was also slightly curved; a short tube connecting the flask atmosphere to vacuum (Figure 1B, 6); and another short tube leading to a 10-psi pressure relief valve (Figure 1B, 5). A 1-inch, magnetic stirring bar was placed in the bottom (Figure 1B, 3). The center neck of the flask was a 2.5-inch o-ring joint (such as on the Ace Glass 8273 adapter), designed to mate with the condenser column and cooling coil assembly above it.

The lower portion of the condenser column consisted of a solution cooling coil (Figure 1B, 4) which was sealed to the center neck of the flask (Figure 1B, 4) using a metal clamp (Figure 1B, 13) around the 2.5-inch o-ring joint (Figure 1B, 9). When in place, the cooling coil extended downward into the solution as far as possible without interfering with the stirring bar. The inlet to the coil was connected to a cold-water source and the outlet to a sink drain. At the top of the cooling coil was a female, 1-inch, standard-taper, ground-glass joint designed to receive the condenser column (Figure 1B, 11) above it. The joint was sealed using a thin film of vacuum grease and held in place with corrosion-free wire springs (7600 Stainless steel Clamp, for joint of size 24/40, ACE Glass catalog number: 7600-25) (Figure 1B, 10).

The condenser column (Figure 1B, 11) consisted of a coiled glass tube that was open to the flask at the bottom and housed inside a water jacket through which cold water was circulated. The top of the coiled tube was connected to a 4-position stopcock which enabled the solution inside the flask to be completely isolated from the atmosphere (position 1), open to the atmosphere (position 2), open to a source of $O_2$-free gas (position 3), or simultaneously open to

159 both the atmosphere and the $O_2$-free gas (position 4). In this last position, the $O_2$-free gas was
160 continuously swept across the top of the condenser column, which maintained an open system at
161 atmospheric pressure inside the flask without permitting $O_2$ to enter back through the column
162 (Figure 1B, 12).

163 The access ports to the condenser flasks in the CALE were connected to a distribution
164 network (Figure 2) through a series of tubes and valves which were operated in concert with each
165 other in various combinations. The valve combinations, together with appropriate settings for the
166 stirrer hot plates, were defined to achieve the specific purposes of the apparatus (see the various
167 Configuration definitions in Table 1). The two major inputs for the network were $O_2$-free Ar and
168 vacuum, and within the network the principal activities were to deoxygenate the flask solutions
169 and to transfer them and supernatant solutions into or out of the IRT through the sample port.

170 The CALE (Figure 2) was prepared with deoxygenated 1 M and 0.005 M solutions of
171 NaCl in flasks 1 and 2, respectively, and with deoxygenated DI water in flask 4. The solution in
172 flask 3 varied depending on the needs of the experiment. For example, when testing the nitrate
173 reduction capacity of a redox-modified smectite (Su et al., 2012), flask 3 was filled with 0.001 M
174 $NaNO_3$. To fill the respective flasks, a volume of 1 to 2 L of the desired solution was prepared in
175 an Erlenmeyer flask or beaker and then connected to the sample port of the CALE through a
176 flexible tube which replaced the needle. Configuration I (Table 1) was then employed to siphon
177 the desired solution into its flask. The solution was then deoxygenated by changing to
178 Configuration II, boiling for 1 h with $O_2$-free gas flowing, then cooled to room temperature using
179 Configuration III in which cold water was circulated through the cooling coil to effect rapid
180 cooling of the solution after the de-oxygenation heating cycle. This cooling coil is not required,
181 but shortens the time needed for cooling. Supernatant solution was removed from the IRT using

Configuration V (supernatant discarded or saved), and the selected deoxygenated solution was transferred to the sample IRT using Configuration VI. Configuration IV was the regime used to store solutions in all flasks after de-oxygenation or when in standby status. Other actions were also possible as defined by the other Configurations (Table 1).

***Preparation stages of the Fe-reduced smectite***

To test the effectiveness of these apparatus in protecting the sample from atmospheric $O_2$, a 50-mg portion of freeze-dried Na-SWa-1 was dispersed in 20 mL of DI water inside the IRT by shaking overnight, then 10 mL of C-B buffer solution was added, the septum-sealing cap re-installed, and the suspension mixed thoroughly using a vortex mixer. This process is referenced JWS Here hereafter as "resuspension in C-B buffer" (Stages 1 and 5, Figure 1).

Reduction of structural Fe in the sample was accomplished by first preheating the sample to 70°C in a water bath, then inserting two needles through the septum. The first needle (6 inch, 22 gauge, deflected point) supplied $O_2$-free Ar gas to the bottom of the dispersion and the rising bubbles effected continuous gentle mixing; the second (2 inch, 22 gauge, deflected point) served as a vent to allow Ar to escape from the head space above the dispersion without permitting a back flow of atmospheric $O_2$. The deflected point design was chosen in order to minimize coring of the septum by the needle. Once the gas flow was established in the IRT and the sample was at temperature, the septum-sealing cap was removed briefly while 200 mg of granular sodium dithionite was added, then the cap was re-installed. The reduction reaction proceeded for 4 h at 70 °C, after which the tubes were removed and cooled at room temperature for 1 h with the Ar gas continuing to purge the vessel. This time and temperature were selected because Komadel et al. (1990) found these to be effective in achieving maximum reduction of the structural Fe.

Lesser levels of reduction can be achieved by decreasing the time (Lee et al, 2006; Ribeiro et al., 2009), temperature (Komadel et al., 1990), and/or amount of sodium dithionite added (Komadel et al., 1990).

In one experiment, the reduction process was monitored continuously by UV-Vis spectroscopy by using a modified version of the IRT (Figure 3) in conjunction with a quartz flow cell (Cary Q6, Part No. 6610015200) and a peristaltic pump (Masterflex® C/L® Dual-Channel Variable-Speed Tubing Pump, model R-77120-52). The modified IRT (Figure 3) consisted of a 50-mL glass centrifuge tube with the same screw cap as in Figure 1. The body of this centrifuge tube was enclosed inside a water jacket (Figure 5) which was connected to Tygon LFL tubing (Tube ID 2.79 mm, R-96429-48) for water and sample circulation by the dual-channel peristaltic pump. The glass reaction vessel (Figure 3, 5) was closed at the top using the septum-sealing cap (Figure 3, 1). Two needles were then inserted into the vessel (Figure 3, 6 and 7) to establish an open system while maintaining an inert atmosphere, as described for sample reduction above. The water circulating in the jacket was taken from the water bath at 70 °C, and the sample was circulated through a quartz flow cell located in the beam path of the spectrophotometer. After about 10 min of temperature equilibration of the Na-SWa-1 suspension with the water bath, a reference spectrum was obtained from 200 to 800 nm, then the wavelength was fixed at 730 nm and continuous recording of the absorbance value at this wavelength was begun. The screw cap was opened and approximately 200 mg of sodium dithionite was added. At selected intervals, the spectrum was scanned from 200 to 800 nm then returned to a fixed value of 730 nm.

***Removing excess salts from redox-activated* Na-SWa-1**

Converting samples into suitable forms for analysis usually requires the removal of reaction products and other solutes (e.g. citrate, bicarbonate, excess NaCl, dithionite degradation

products, etc.) associated with the reduction reaction, and minimizing the concentration of solutes in the outer solution of the dispersion. This was accomplished by centrifuge washing (Sorvall Dupont Model RC4-M Plus centrifuge with SS-34 rotor) at 1400 x g (3000 rpm) **[Martin, are these numbers correct? Shouldn't the g force be greater than the rpm?]**.

If the supernatant remained cloudy, the g force was doubled. During this washing procedure, supernatant solutions were decanted and discarded using Configuration V of the CALE, as described above, with the first needle (6-inch, 22 gauge, deflected point) inserted into the supernatant to a point just above the sediment (down location, Table 1) and the second needle (6-inch, 22 gauge, deflected point) inserted into the head space (up location, Table 1). Supernatant was withdrawn by vacuum into the discard container and a positive pressure was maintained inside the vessel by Ar addition. New solution was then added using Configuration VI. Sample re-dispersion was accomplished by vortex mixing, vigorous shaking, and/or bath sonication for 15 min, depending on the agglomeration in the sample. Vortexing while slowly refilling helps the redispersion process. These steps allowed the exchange of solutions in the IRT without exposing the redox-activated Na-SWa-1 to atmospheric $O_2$. The initial supernatant solution was replaced with 1 M NaCl from flask 1; followed by three to five more washings with 0.005 M NaCl from flask 2. These washing steps used 1400 x g for 20 min. After decanting in the last washing step, another solution (from flask 3 or 4) or nothing was added, depending on the experiment.

***Glove-box handling and storage***

A glove box with antechamber (Vacuum Atmospheres Model HE-4), equipped with a Pedratol automatic pressure-control system and a dry train through which the $N_2$ or Ar atmosphere was continuously circulated to remove $H_2O$ and $O_2$ was used to store some samples,

to transfer samples to some types of holders, and to freeze dry the sample under an inert atmosphere (Figure 4).

Freeze drying capability under an inert atmosphere was added to the inside of the glove box by installing a vacuum valve and tubing and a thermoelectric cold plate (model CP-2, Thermoelectric Unlimited, Inc., Wilmington, Delaware) The cold plate was equipped to hold and freeze the contents of up to 4 IRTs simultaneously (Figure 4, 2). A special cap was manufactured to replace the sealing-cap assembly and to connect the IRT to the vacuum line (Figure 4, 4). When in place, this cap securely isolated the evacuated IRT atmosphere from the glove-box atmosphere, thus preventing the applied vacuum from lowering the pressure inside the glove box. Once the sample was securely frozen, the vacuum valve was opened and the atmosphere within the IRT was then continuously evacuated to sublime the frozen $H_2O$ from the sample gel.

***Stages for Analyzing Reduced Na-SWa-1***

Reduced samples were then investigated at certain stages of the chemical reduction and washing processes in order to assess the reliability of the inert-atmosphere handling methods. These stages were (Figure 1):

Stage 0: Freeze-dried, Na-saturated, <2-µm particle-size fraction of ferruginous smectite (labeled Na-SWa-1).

Stage 1: Freeze-dried, Na-SWa-1 from Stage 0, resuspended in C-B buffer.

Stage 2: Freshly reduced Na-SWa-1 in suspension prior to any washing or further treatment.

Stage 3: A compressed gel obtained by centrifuging the suspension in Stage 2 at 35,000 x g (20,000 rpm) and decanting the supernatant.

Stage 4: Freeze-dried form of the reduced, unwashed sample from Stage 3, using the freeze dryer inside the glove box.

Stage 5: Resuspension of freeze-dried sample from Stage 4 with C-B buffer.

Stage 6: Washed (5X) samples from Stage 2 (A, washed with CALE; B, washed without CALE) or Stage 5 (C, washed with CALE). Washing was with a 5 mM NaCl or $H_2O$ solution and left in suspension after the final wash.

Stage 7A, 7B, 7C: A compressed gel obtained by centrifuging the respective suspensions in Stage 6 at 35,000 x g (20,000 rpm) and decanting the supernatant.

Stage 8A, 8B, 8C: Freeze-dried form of the reduced, compressed gels from Stages 7A, 7B, 7C, respectively..

***Chemical Analysis for Structural Fe Reduction***

The reduced state (Fe(II) content) of the smectite was determined in either the gel or freeze-dried state using the 1,10-phenanthroline method (Komadel and Stucki, 1988). The septum-sealing cap was removed from the IRT containing the sample in either Stage 4 or 5, then 12 mL of 3.6 N $H_2SO_4$, 2 mL of 10% (w/v) 1,10-phenanthroline in 95 % ethanol, and 1 mL of 48 % HF were added immediately, in that order, under red lights. These solutions were not purged of $O_2$ because the low pH prevented oxidation of the tris(1,10-phenanthroline)Fe(II) complex. The open tubes were heated for 30 min in a boiling water bath and cooled for 15

minutes at room temperature. To remove excess $F^-$, 10 mL of 5% (w/v) $H_3BO_3$ was added. The solutions were then diluted as prescribed by the method (Stucki, 1981; Stucki and Anderson, 1981; Komadel and Stucki, 1988), and analyzed for Fe(II) at 510 nm (Varian Cary 5E UV-Vis-NIR spectrophotometer). Total Fe was measured after exposing the diluted samples to ultra-violet light for 2 h. For freeze-dried samples (Stage 5), the amount of Fe(II) and total Fe were calculated from the Beer-Lambert Law, *viz.*,

$$A_i = \epsilon_i l C_i \quad [1]$$

where $i$ refers to either Fe(II) or total Fe, $A_i$ is the absorbance at 510 nm of either the Fe(II) or total Fe solution, $\epsilon_i$ is the absorbance of the 1,10-phenanthroline Fe(II) complex in the Fe(II) or total Fe solution (0.1836 and 0.1921 g/µg-cm for Fe(II) and total Fe, respectively), $l$ is the path length (exactly 1.00 cm for all analyses), and $C_i$ is the concentration of Fe(II) or total Fe (µg/g) in the analyte solution.

For samples analyzed while in the undried gel state (Stage 4 or 7), the exact amount of starting material was unknown, so only the ratio of Fe(II)/Total Fe $\left({C_{Fe(II)}}/{C_{Total}}\right)$ could be determined, which was calculated from Equation 2, using the ratios of absorbance values $\left({A_{Fe(II)}}/{A_{Total}}\right)$ from Equation 1.

$$\frac{C_{Fe(II)}}{C_{Total}} = \frac{A_{Fe(II)}\epsilon_{Total}}{A_{Total}\epsilon_{Fe(II)}} \quad [2]$$

***Mössbauer Spectroscopy***

Mössbauer spectra were collected using a Web Research, Inc. (Edina, Minnesota), spectrometer equipped with a Janis model SHI-850-5 (Janis, Inc., Wilmington, Massachusetts) closed cycle cryostat and operating in triangle waveform mode with a 50 mCi $^{57}$Co source dispersed as 10 wt.% in a thin Rh foil, obtained from Ritverc, Inc (St. Petersburg, Russia). The velocity scale was calibrated using the magnetic hyperfine field ($B_{hf}$) of a 7 μm thick α-Fe foil at the sample temperature. To estimate the correct value for $B_{hf}$ at 77 K, Mössbauer spectra of the alpha Fe were collected at 4, 77, and 298 K, then the number of channels separating peaks 1 and 6 were plotted as a function of temperature. Values for $B_{hf}$ for α-Fe at 4 and 298 K were determined by Violet and Pipkorn (1971) to be 33.9 *T* and 33.1 *T*, respectively. The corresponding value at 77 K was found to be the same as at 4 K, i.e. 33.9 *T*, so this value was used to calibrate the velocity scale.

## RESULTS AND DISCUSSION

The effectiveness of these methods and apparatus for preserving the oxidation state during the preparation, handling, and storage of redox-sensitive ferruginous smectite samples was tested in several ways. First, the color was monitored, both by eye and spectroscopically. Second, the structural Fe(II) content was measured chemically, either gravimetrically or as a ratio of Fe(II) to total Fe, using the 1,10-phenanthroline method. And third, the extent of reduction and structural alteration was assessed using Mössbauer spectroscopy with the sample at 77 K.

### Color

Unreduced and reduced Na-SWa-1 samples differed markedly in their color, as is well known (Roth and Tullock, 1973; Rozenson and Heller-Kallai, 1976; Stucki and Roth, 1977; Russell *et al.*, 1979; Stucki et al., 1984; Komadel and Stucki, 1990; Merola et al., 2007; Stucki,

1988, 2006, 2013). The yellow-brown color of oxidized or unaltered CB-buffered Na-SWa-1 suspensions turned rapidly to green-yellow upon addition of sodium dithionite powder. With time, the color changed progressively to green, blue-green, blue, blue-grey, and light grey, which colors represented increasing structural Fe(II) content. Komadel and Stucki (1990) found that the progression of these color changes, and hence of reduction, was enhanced by higher temperature (70 °C) and by longer reaction time. They also found that the ideal dithionite:smectite ratio was on the order of about 5:1 and that no further color change was observed after 4 h of the reduction treatment at 70 °C. The grey color of the suspension, which occurs only in Fe-rich smectites, indicated that reduction was finished.

These colors were also measured spectroscopically by monitoring the intervalence electron transfer transition that occurs at about 730 nm in Fe-rich smectite (Figure 5). The general trend in absorbance values with time of reduction was similar to that observed by Komadel and Stucki (1990) for ferruginous smectite. The absorbance values at 0 and 4 h represented the band intensities for the unaltered and maximum-reduced Na-SWa-1. The sample at 4 h corresponded to samples prepared in the water bath at Stages 2 and 3. Because dithionite was still in these suspensions, no attempt was made to analyze them chemically for Fe(II) and total Fe, but they were analyzed by Mössbauer spectroscopy at 77 K (see below).

$f_{295}$ were 0.821–0.917 for $Fe^{3+}$ and 0.662–0.743 for Fe2+

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Mössbauer spectroscopy of phyllosilicates: effects of fitting models on recoil-free fractions and redox ratios *Clay Minerals, March 2008, v. 43, p. 3-33, published online 1 April 2008,*

Celadonite (from deGrave and van Alboom, 1991) gives f2 = 0.892 and f3 = 0.935, for a f2/f3 ratio of 0.954

C2/C1 = k * A2/A3 where k=f3/f2

A1/A2 = C N1/N2 where C = a1 b1 f1/a2 b2 f2 (see equation 3 and 4 of Dyar et al.)

Generally stated, f3+ > f2+

Dyar found f3 in nontronite and SWa1 to be 0.916 to 0.919, but no f2 data.

Biotite gave f3 = 0.942 and f2 = 0.871 for a f2/f3 = 0.924

**Mössbauer Spectroscopy**

Na-SWa-1 reduced and treated up to Stage 3 was transferred to the Mössbauer sample holder while inside the glove box, then the Mössbauer spectrum was obtained at 77 K (Figure 6). Results revealed a sharp doublet characteristic of Fe(II) that accounted for 92.2% of the total spectral area, with a small feature for Fe(III) accounting for the remaining area (7.8%), indicating that the sample was highly reduced. This result is very close to the chemical analysis, which found the Fe(II) content to be 91.129 % (±0.477 %) of Total Fe (Table 2, Figure y). This appears to be the maximum reduction level possible for ferruginous smectite.

After washing with the CALE (Stage 4), the spectrum (Figure 5) revealed that the Fe(III) content increased only slightly to ?? % amount of Fe(III), and after freeze drying (Stage 5), the spectrum had slightly more Fe(III) than before freeze drying. The corresponding values obtained

by chemical analysis were % and % (Figure y). The Mössbauer spectrum of a sample washed without removing $O_2$ from the wash solutions (Stage 3b) revealed an increase in Fe(III) content of about ?? %. These Mössbauer spectra were clear indicators that the methods employed to prevent reoxidation were highly effective.

Stucki and co-workers (1984) observed a much lower level of Fe(III) conversion to Fe(II) in C-B buffered Garfield nontronite suspensions at 25 °C than at 70 °C. Beside temperature, they also studied the influence of sodium dithionite vs. clay loading and the reaction time on Fe reduction level in Garfield nontronite. They reached the higher transformation of Fe from ferric to ferrous oxidation state at 70 °C for 168 hours with loading 600 mg of sodium dithionite and 100 mg of the clay, respectively. In spite of strong conditions, the ratio of Fe(II):total Fe was ~ 0.779 which was maybe caused by insufficient handling with the samples. The efficiency of our developed procedure and improvement of presented a washing CALE apparatus for iron reduced clay samples is in Table 1. Analysis of solid phases by 1, 10-phenanthroline method performed 10 times provided a high conversion level for 91.129 ± 0.477 % of Total Fe %, due to high precise handling during reduce iron clay preparation.

## CONCLUSIONS

Suspensions of Na-SWa-1 were reacted with sodium dithionite and buffered by sodium citrate-sodium bicarbonate solution at 70 °C for 4 hours. Changes in color of unreacted and reacted suspensions provided the clear evidence that iron in the clay structure was reduced. An iron reductions under oxygen free conditions were performed with minimum Fe(II) back re-oxidations to Fe(III). The proof of this argument was obtained from Fe(II) and total Fe determination. Statistically, a transformation of the Fe(III) reduction to Fe(II) equals to 91.129 ± 0.477 % of Total Fe %. A controlled atmosphere liquid exchange (CALE) apparatus was

introduced and described in detail. Suitability of CALE apparatus for air sensitive iron clay redox systems was demonstrated with success. It was effective in reaction products removal and maintaining of high Fe reduction.

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511

## TABLE AND FIGURE CAPTIONS

Table 1. The various configuration definitions for specific purposes of the CALE apparatus.

Figure 1. Schematic drawing of the CALE apparatus.

Figure 2. The details of the polycarbonate centrifugation tube (A) and the glass part (B) of a CALE apparatus.

Figure 3. Schematic drawing of the Glove Box and Freeze Drying procedure.

Figure 4. The water jacked glass inert-atmosphere reaction tube (IRT).

Figure 5. A comparative iron analysis of the clay (SWa-1) samples washed with and without CALE apparatus after dithionite reduction

# Table

| Configuration | A | B | C | D | E | F | G | H | I | J | K | L | M | N | O | P | Q | R |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| I. Filling flask with solution | Off/NA | Soln | 1 | 1c | On | Off | 1 | 3 | On | Off | Off | Off | Off | Off | Down | NA | NA | NA |
| | Off/NA | Soln | 2 | 2c | On | Off | 2 | 3 | On | Off | Off | Off | Off | Off | Down | NA | NA | NA |
| | Off/NA | Soln | 3 | 3c | On | Off | 3 | 3 | On | Off | Off | Off | Off | Off | Down | NA | NA | NA |
| | Off/NA | Soln | 4 | 4c | On | Off | 4 | 3 | On | Off | Off | Off | Off | Off | Down | NA | NA | NA |
| II.Deoxygenation of solution | 1# | Off | 1/NA | 1a# | Off | Off | 1~/NA | 1 | Off/On~ | 1# | On | On | On | Off | NA | NA | NA | NA |
| | 2# | Off | 2/NA | 2a# | Off | Off | 2~/NA | 1 | Off/On~ | 2# | On | On | On | Off | NA | NA | NA | NA |
| | 3# | Off | 3/NA | 3a# | Off | Off | 3~/NA | 1 | Off/On~ | 3# | On | On | On | Off | NA | NA | NA | NA |
| | 4# | Off | 4/NA | 4a# | Off | Off | 4~/NA | 1 | Off/On~ | 4# | On | On | On | Off | NA | NA | NA | NA |
| III. Cool deoxygenated solution | 1# | Off | 1/NA | 1a# | Off | Off | 1/NA | 1 | Off | 1 | Off | On | On | On | NA | NA | NA | NA |
| | 2# | Off | 2/NA | 2a# | Off | Off | 2/NA | 1 | Off | 2 | Off | On | On | On | NA | NA | NA | NA |
| | 3# | Off | 3/NA | 3a# | Off | Off | 3/NA | 1 | Off | 3 | Off | On | On | On | NA | NA | NA | NA |
| | 4# | Off | 4/NA | 4a# | Off | Off | 4/NA | 1 | Off | 4 | Off | On | On | On | NA | NA | NA | NA |
| IV. Store solution | Off/NA | Off | 1/NA | 1b | On | Off | 1 | 3 | On | Off | Off | Off | Off | Off | NA | NA | NA | NA |
| | Off/NA | Off | 2/NA | 2b | On | Off | 2 | 3 | On | Off | Off | Off | Off | Off | NA | NA | NA | NA |
| | Off/NA | Off | 3/NA | 3b | On | Off | 3 | 3 | On | Off | Off | Off | Off | Off | NA | NA | NA | NA |
| | Off/NA | Off | 4/NA | 4b | On | Off | 4 | 3 | On | Off | Off | Off | Off | Off | NA | NA | NA | NA |
| V. Decant and discard/save | Off/NA | Argon | NA | Off/NA | On | Discard | NA | 3/NA | Off | Off | Off | Off | Off | Off | Up | Down | NA | NA |
| | Off/NA | Argon | NA | Off/NA | On | Save | NA | 3/NA | Off | Off | Off | Off | Off | Off | Up | Down | Up | Up |
| VI. Transport solution to the sample | Off/NA | Soln | 1 | 1c | On | either | 1/NA | 3 | Off | 1* | Off | Off | Off | Off | Up | Up | NA | NA |
| | Off/NA | Soln | 2 | 2c | On | either | 2/NA | 3 | Off | 2* | Off | Off | Off | Off | Up | Up | NA | NA |
| | Off/NA | Soln | 3 | 3c | On | either | 3/NA | 3 | Off | 3* | Off | Off | Off | Off | Up | Up | NA | NA |
| | Off/NA | Soln | 4 | 4c | On | either | 4/NA | 3 | Off | 4* | Off | Off | Off | Off | Up | Up | NA | NA |

#set an adequate flow rate

* use this option in the case of unsafficient solution flow rate

~use this option in the case of the solution outflow through the valve at the top of the condenser

A: Condenser sweep
B: Sample port
C: Solution selection
D: Feed to sample port – position a=Ar; position b=Off; position c=flask solution
E: Main vacuum On or Off
F: Decant selector, save or discard
G: Vacuum to flask: selector
H: Valve at the top of the condenser; position 1=all legs open; position 2=flask vented to right; position 3=flask closed
I: Main vacuum to flask (on or off)
J: Solution purge
K: Hot Plate
L: Stir Bar
M: Condenser
N: Cool Coil
O: Needle 1, (from "Sample Port")
P: Needle 2, (to "Decant Selector")
Q: Needle 3, (from "Decant Selector")
R: Needle 4, (to "Vacuum")

## Figures

Figure 1.

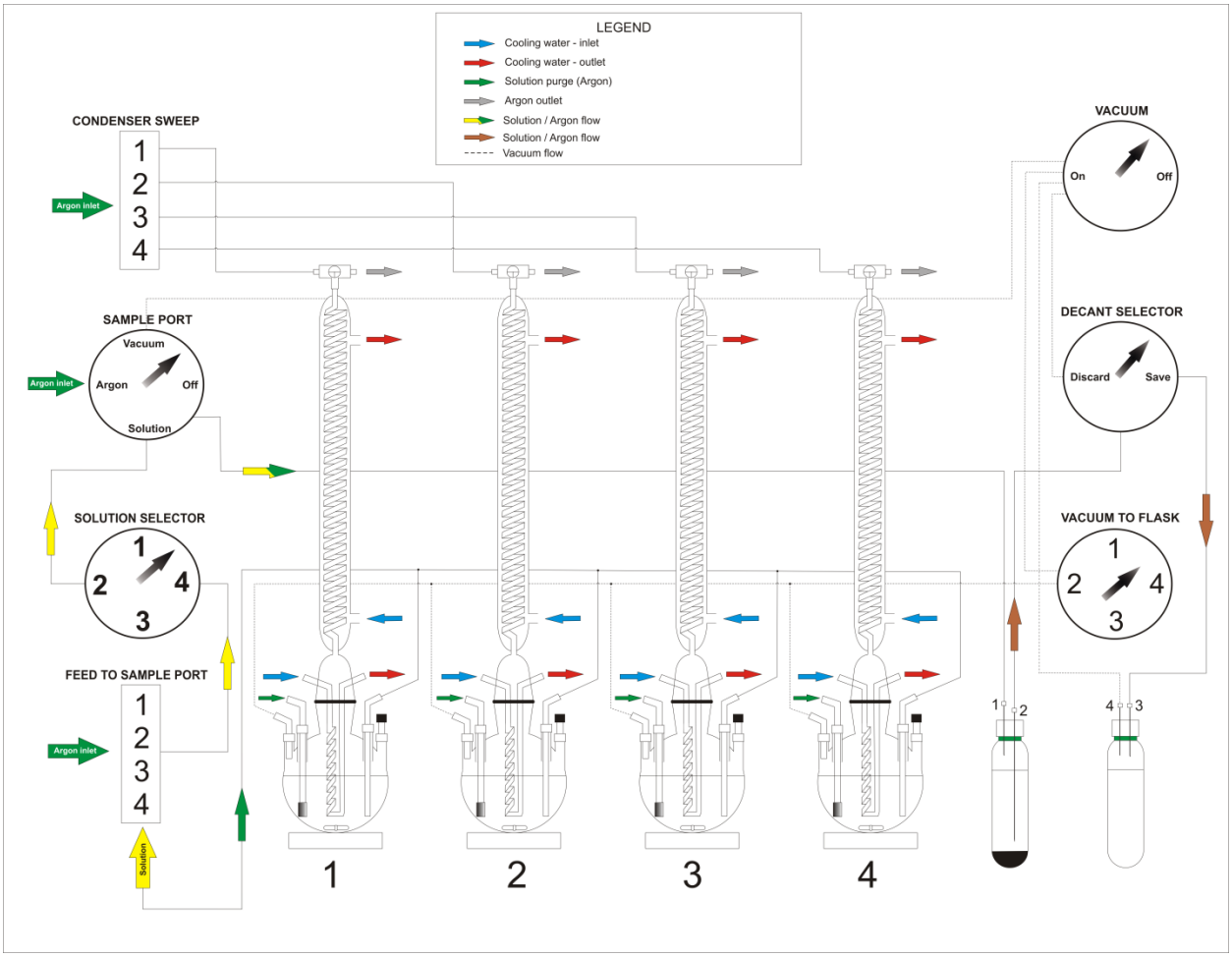

Figure 2.

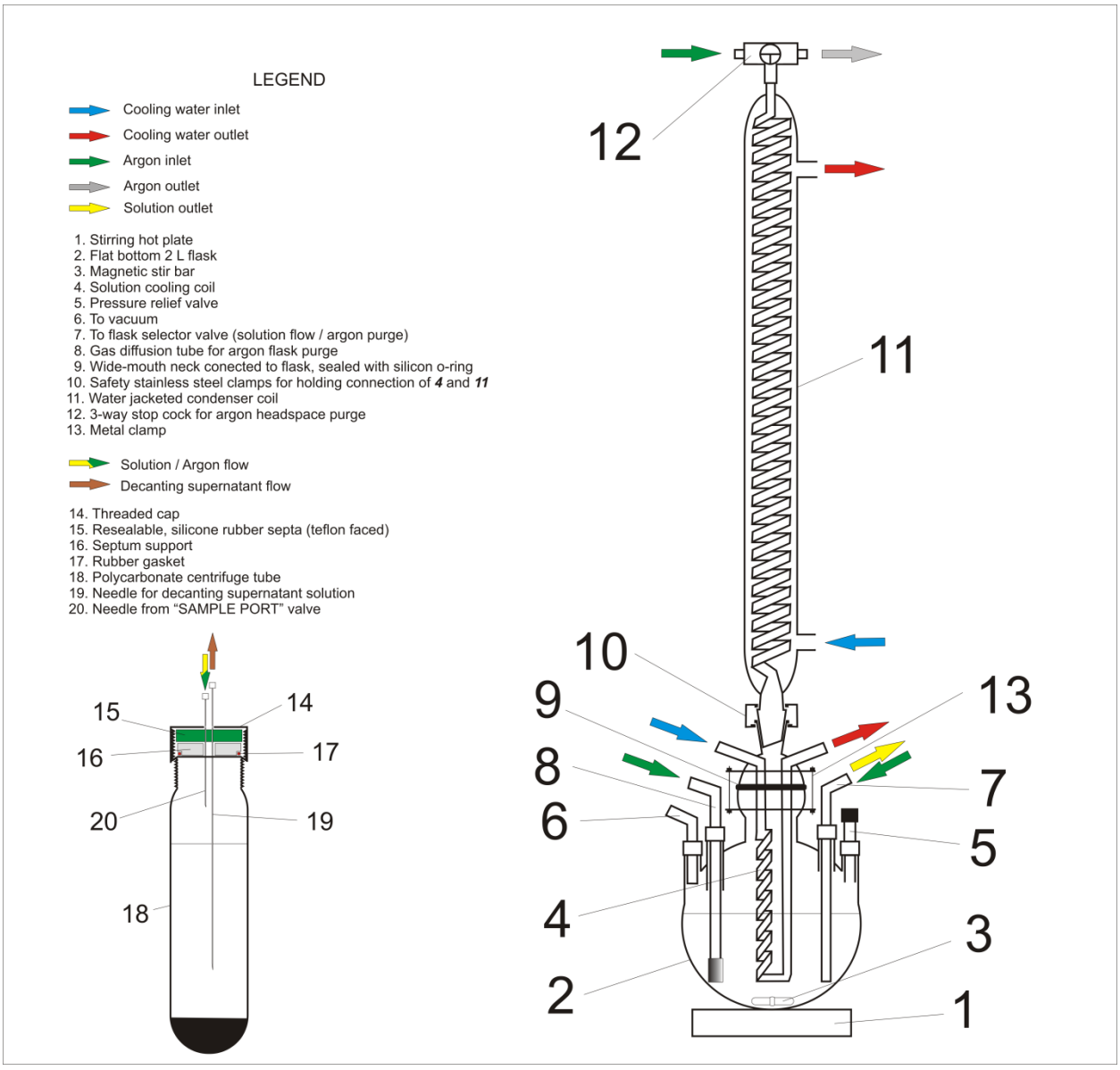
LEGEND
Cooling water inlet
Cooling water outlet
Argon inlet
Argon outlet
Solution outlet
1. Stirring hot plate
2. Flat bottom 2 L flask
3. Magnetic stir bar
4. Solution cooling coil
5. Pressure relief valve
6. To vacuum
7. To flask selector valve (solution flow / argon purge)
8. Gas diffusion tube for argon flask purge
9. Wide-mouth neck conected to flask, sealed with silicon o-ring
10. Safety stainless steel clamps for holding connection of 4 and 11
11. Water jacketed condenser coil
12. 3-way stop cock for argon headspace purge
13. Metal clamp
Solution / Argon flow
Decanting supernatant flow
14. Threaded cap
15. Resealable, silicone rubber septa (teflon faced)
16. Septum support
17. Rubber gasket
18. Polycarbonate centrifuge tube
19. Needle for decanting supernatant solution
20. Needle from "SAMPLE PORT" valve
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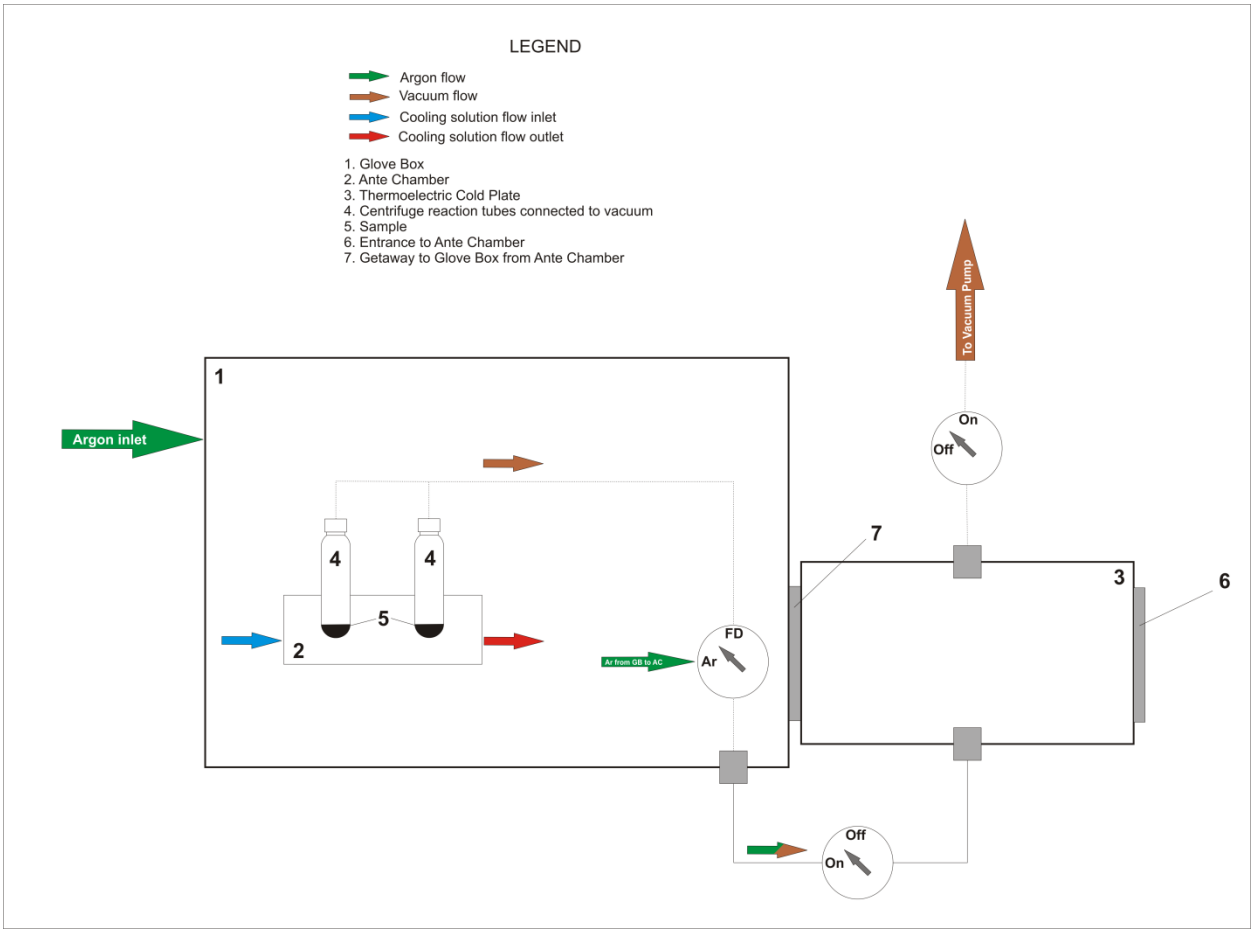
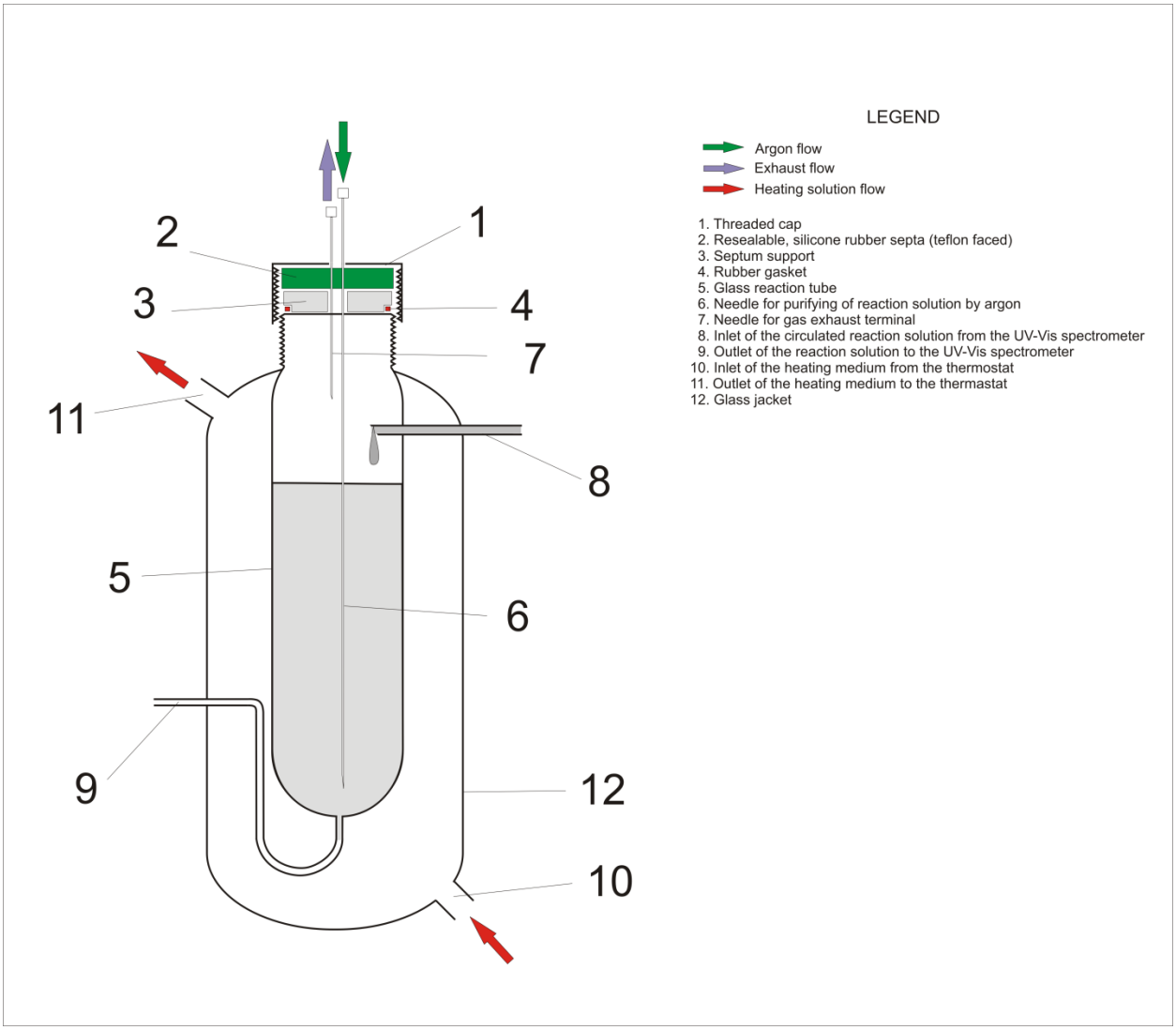
Figure 3.


LEGEND
Argon flow
Vacuum flow
Cooling solution flow inlet
Cooling solution flow outlet
1. Glove Box
2. Ante Chamber
3. Thermoelectric Cold Plate
4. Centrifuge reaction tubes connected to vacuum
5. Sample
6. Entrance to Ante Chamber
7. Getaway to Glove Box from Ante Chamber
To Vacuum Pump
On
Off
Argon inlet
1
4
4
5
2
7
3
6
FD
Ar
Ar from GB to AC
Off
On

549 Figure 4.

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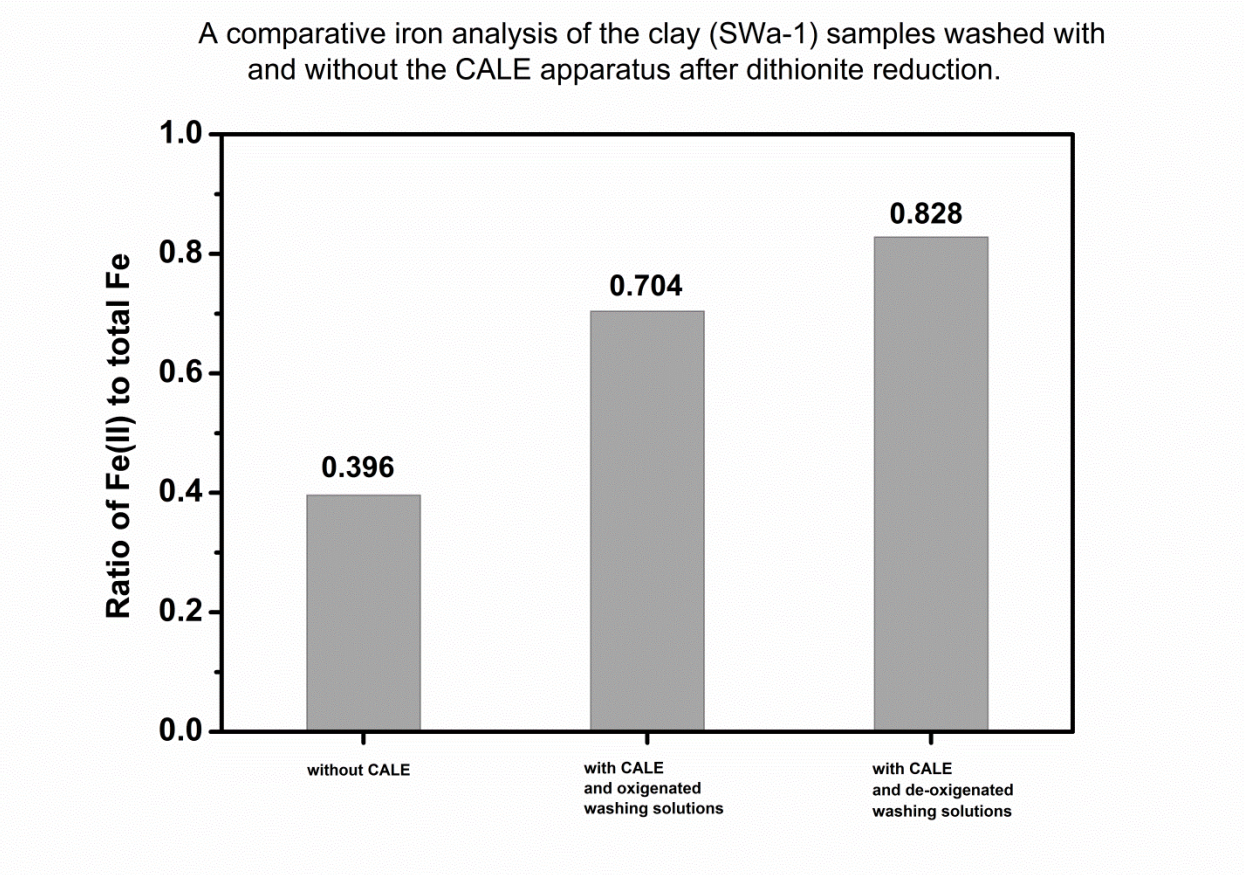
556 Figure 5.

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598 *Journal of Applied Physics,* **42,** 4339-&


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With the cooling coil 8 and condenser 10 removed from the flask, the washing solution was poured into the flask through the wide mouth 9, then 8 and 10 were joined together by a standard-taper, ground-glass joint and 8 was attached to the wide-mouth joint 9 of the flask using an o-ring and metal clamp. Cold tap water was circulated continuously through the outer jacket of condenser column 10 and $O_2$-free Ar gas was swept through the 3-way valve 11 across the top of column 10, with the valve also open to the flask. Oxygen-free Ar was then admitted into the solution through gas dispersion tube 5 by the proper setting of the 3-way valve A (Figure 6, "Feed to sample port") was opened so Ar sweep gas solution was then deoxygenated by then the cooling coil on a stirring hot plate (Fig.3, B-2) equipped with a magnetic stir bar (Fig.3, B-3). A condenser coil (Fig.3, B-8) is attached to the central neck of each flask, which is used for rapid cooling of the solution after boiling. Above the whole flask at the central neck is a water jacketed condenser column for total reflux of boiling solution (Fig.3, B-9). Oxygen-free argon inlet controlled by 3-way valve "Feed to sample port" and loaded to the one of four little threaded necks (Fig.3, B-5) passed through solution via gas dispersion tube and solutions were boiled approximately 1 hour with stirring, because the solubility of the gases in the water decreases with increasing temperature. The 3-way valve (Fig.3, B-10) at the top of reflux cooler was opened to secure outlet exhaust and avoid increase of pressure in the flasks. The stopcock was closed after the solutions had been cooled and some solution was passed via solution outlet (Fig.3, B-6). The little threaded neck (Fig.3, B-7) is equipped with the pressure relief valve (10 PSI) for safety and during all operations of the CALE apparatus must be covered by the transparent safety shield (Fig.1).

The IRT containing the freshly reduced, cooled Na-SWa-1 was centrifuged (Sorvall Dupont Model RC4-M Plus centrifuge with SS-33 rotor) at 5000 rpm which corresponds to 3000

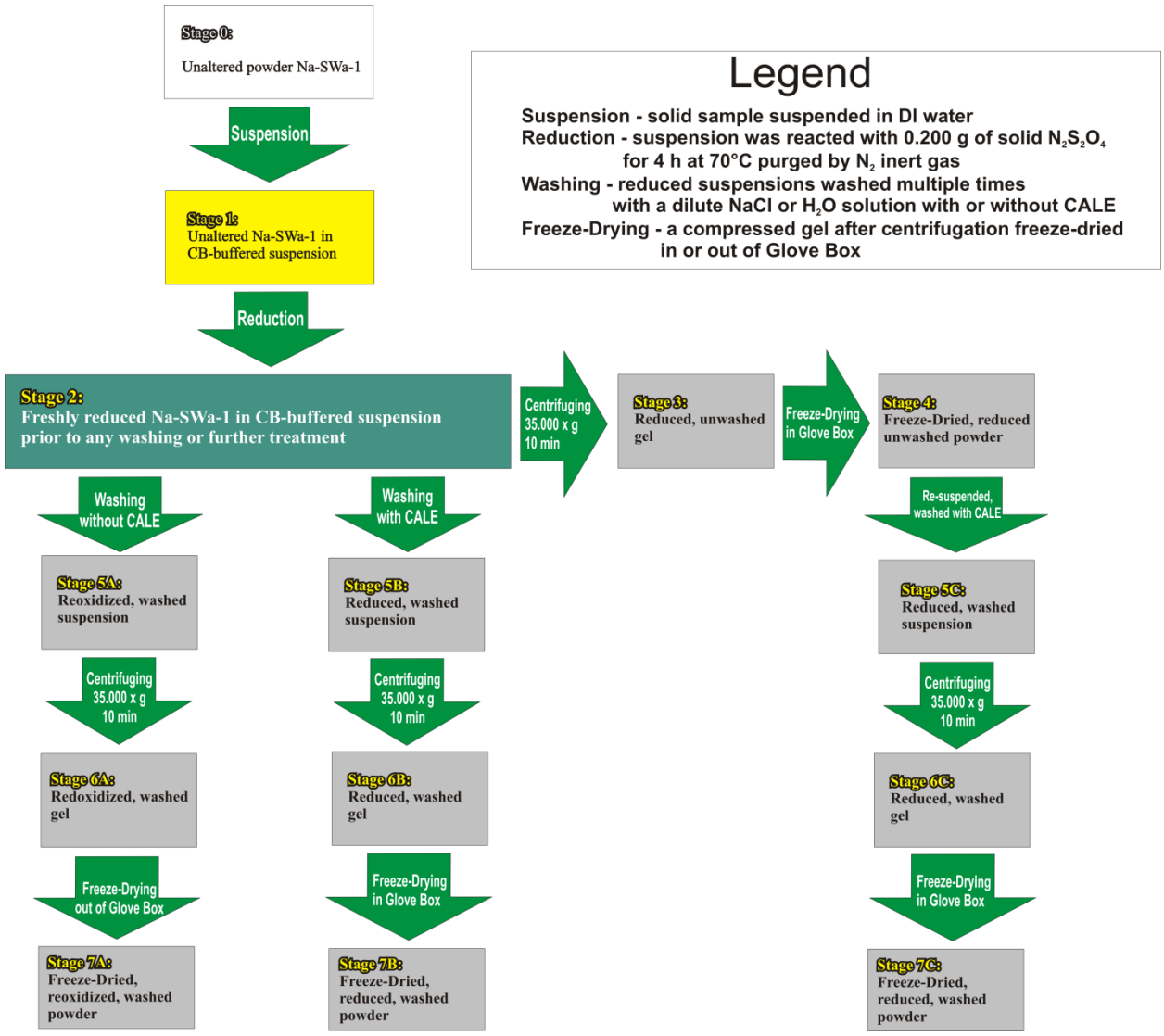
x g (Martin, please confirm this rpm and calculate the corresponding g force), then the supernatant solution was decanted and replaced with 1 M NaCl from flask 1 (Figure 1 and Figure 4). This washing step was repeated three to five more times using 0.005 M NaCl from flask 2. After the last washing step with NaCl, DI water was added from flask 3. The figure 3 shows details of the reaction centrifugation tube (A) and the glass part of CALE apparatus (B).

Prior to washing steps the samples were centrifuged for 15 minutes at 1400 x g with the SS-34 rotor on the Dupont Sorvall Model RC 5C plus. In the case of insufficient dispersion presence (supernatant occurs cloudy) the double centrifuge speed is needed. When the supernatants were clear and washing solutions properly deoxygenated, the decanting of the supernatants was performed by CALE without exposure to the atmosphere. Two septum penetration needles are inserted into to the centrifugation tubes (Fig.3, A-11) via silicon rubber septa. The needles were designed with the tip in the side wall for penetration without coring a hole. The needle (Fig.3, A-12) lowered into the clear supernatant solution one cm above sediment in the bottom for decanting the supernatant solution, which feeds through the Tygon tube into a large neck filter flask under vacuum. The needle (Fig.3, A-13) inserted about one cm through the septa cap. This needle is attached to a 4-way valve “SAMPLE PORT”. In position “OFF” the needle is closed. In position “Argon“ is used for pumping the $O_2$-free Argon into the centrifuge tube to enhance the pressure gradient for the supernatant solutions removal support and to prevent negative pressures inside the tube and feeding a new washing solution from selected bottom rounded flasks (Fig.3, B-1). In position “VACUUM” the needle can be used as a vacuum needle. In position “SOLUTION” the needle is connected to a 4-way valve “SOLUTION SELECTOR” which allows inflow of the fresh deoxygenated solution from the selected flasks (Fig.1).

Once “the needle to vacuum” (Fig.3, A-12) is inserted into the centrifugation tube through the green silicon septa the supernatant is vacuumed and the pressure in the reaction tube getting low. This deficit must be compensated by incoming argon provided by “the needle from the flask” (Fig.3, A-13), connected to the 4-way valve “SAMPLE PORT” which is set to “Argon” position (Fig.1 and Fig.4). The supernatant is removed and fresh deoxygenated washing solution of 1 mol.dm$^{-3}$ NaCl is not added, leaving a small amount of reduced clay in an atmosphere of argon within the tube and “the needle to vacuum” retrieved 1 cm under silicon septum for the fresh washing solution income enhancing. At need, the extracted solutions can be saved or wasted, the “DECANT SELECTOR” valve in position “SAVE” or “DISCARD”, respectively. The three steps to get washing solutions inside the reaction tubes are needed. “FEED TO SAMPLE PORT” adjusted to “FLASK 1” position, “SOLUTION SELECTOR” in position “1” and “SAMPLE PORT” in position “SOLUTION”, provided the $O_2$-free 1 mol.dm$^{-3}$ NaCl solution flowing to the centrifugation tube. When the required volume of fresh solution is reached, “the needle to vacuum” is released and the “SAMPLE PORT” is switched to “OFF” position (Fig.1 and Fig.4).

Prepared oxygen sensitive redox samples for storage should be placed into the main store-room via antechamber for next use. The first be sure that gateway between the main store and antechamber is closed securely. Then the right door can be opened and samples placed to sliding tray and immediately closed to prevent oxygen penetration into the glove box (Fig.6). Now the glove box should be purged by Nitrogen and Oxygen in antechamber removed. The air from the antechamber will be evacuated by “EVACUATION VALVE” till the vacuum reaches -100 in Hg. When the “EVACUATION VALVE” is closed, the automatic pressure control will start fill the glove box and the red LODs are moved to the left – maximum gas filling. When the

orange LEDs indicate an over-pressure, the “REFILL VALVE” must be opened. The Nitrogen from the main store-room starts taking place in the antechamber and the manometer indicates the pressure increasing till + 30 inHg. The pressure in the glove box must never go to the right. When this limit is reached, the “REFIL VALVE” must be closed and “EVACUATION VALVE” opened. To reach the total oxygen replacement for nitrogen in the glove box, this procedure should be repeated for four times. In case of many samples or material placed in the antechamber, the “refilling & evacuating” cycles must by repeated five times or more, due to larger amount of oxygen trapped inside and risks bringing too much oxygen inside. The common operating conditions inside the glove box, the pressure on the automatic pressure control system is set to + 2 in water (red LEDs). After the purging cycles have been completed, the door between the main store-room and the antechamber can be opened and the samples removed from the sliding tray and deposited inside the main store-room.


Stage 0:
Unaltered powder Na-SWa-1
Suspension
Stage 1:
Unaltered Na-SWa-1 in CB-buffered suspension
Reduction
Legend
Suspension - solid sample suspended in DI water
Reduction - suspension was reacted with 0.200 g of solid $N_2S_2O_4$ for 4 h at 70°C purged by $N_2$ inert gas
Washing - reduced suspensions washed multiple times with a dilute NaCl or $H_2O$ solution with or without CALE
Freeze-Drying - a compressed gel after centrifugation freeze-dried in or out of Glove Box
Stage 2:
Freshly reduced Na-SWa-1 in CB-buffered suspension prior to any washing or further treatment
Centrifuging 35.000 x g 10 min
Stage 3:
Reduced, unwashed gel
Freeze-Drying in Glove Box
Stage 4:
Freeze-Dried, reduced unwashed powder
Washing without CALE
Washing with CALE
Re-suspended, washed with CALE
Stage 5A:
Reoxidized, washed suspension
Stage 5B:
Reduced, washed suspension
Stage 5C:
Reduced, washed suspension
Centrifuging 35.000 x g 10 min
Centrifuging 35.000 x g 10 min
Centrifuging 35.000 x g 10 min
Stage 6A:
Redoxidized, washed gel
Stage 6B:
Reduced, washed gel
Stage 6C:
Reduced, washed gel
Freeze-Drying out of Glove Box
Freeze-Drying in Glove Box
Freeze-Drying in Glove Box
Stage 7A:
Freeze-Dried, reoxidized, washed powder
Stage 7B:
Freeze-Dried, reduced, washed powder
Stage 7C:
Freeze-Dried, reduced, washed powder

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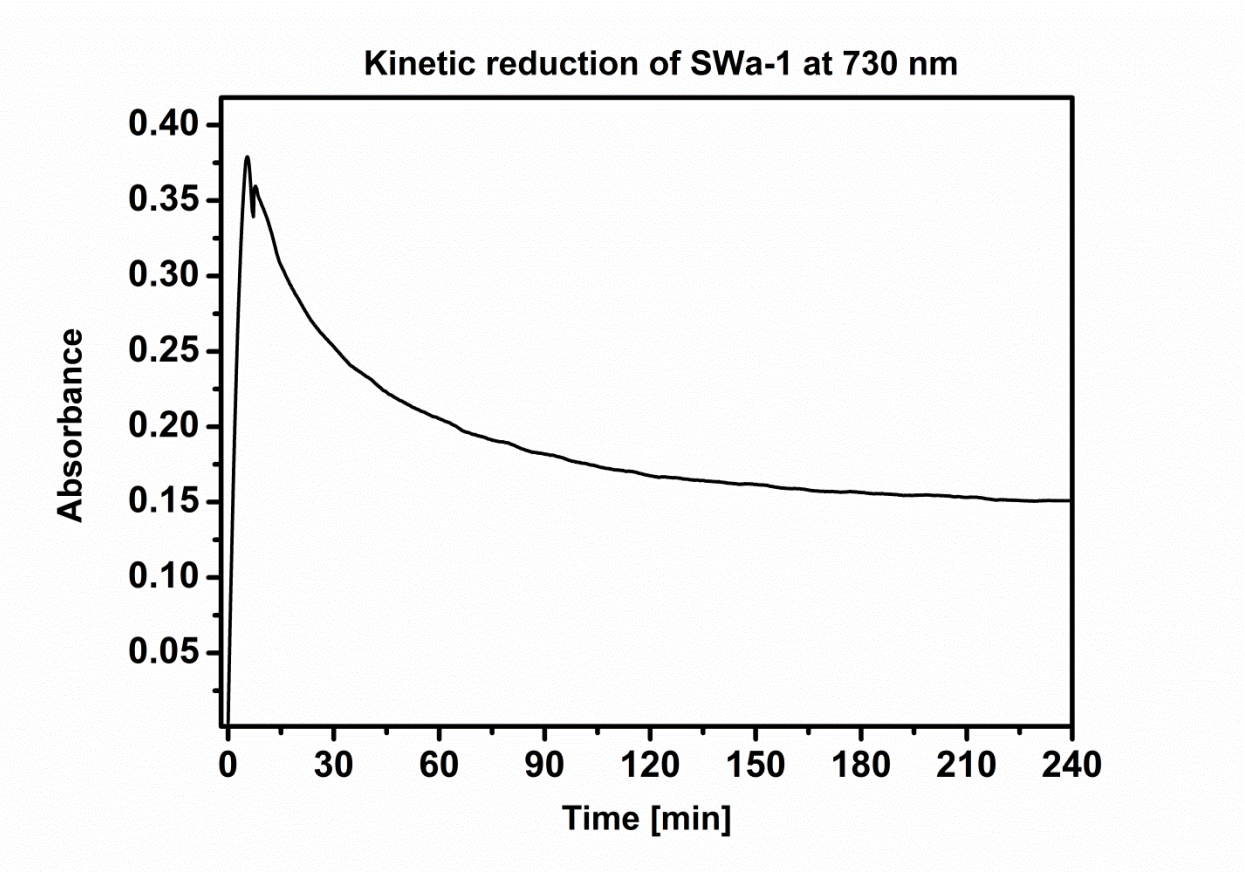


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688 Figure 5. Intervalence electron transfer transition intensity (Absorbance) of structural Fe in
689 SWa-1 with time of exposure to dithionite at 70 °C.

690

691 consisted of three essential features: (1) Several condenser flasks placed on stirrer hot
692 plates in which selected solutions were deoxygenated; (2) a distribution and control network
693 consisting of valves, fittings, and tubing to connect the IRT with selected flasks, decanting
694 circuits, $O_2$-free gas supplies, and vacuum, and to connect the flasks with $O_2$-free gas supplies
695 and vacuum; and (3) a housing in which the flasks were placed to protect the operator from
696 accidental glass breakage and to support the distribution and control network with a comfortable
697 operator interface and work station (Figure 2).

698