



VINTESSENTIAL LABORATORIES FREE & TOTAL SO₂ KITS

Free & Total SO2 Kits by Vintessential Laboratories for Manual Spectrophotometers

These kits are slightly different than the other kits made by Vintessential. The following describes the major differences between the free & total SO₂ kits and other enzymatic kits from Vintessential Laboratories:

1) Semi-Micro Cuvettes are required for these kits. They look like the typical 10 mm cuvettes we sell, but they are filled in with more plastic to make the volume size in the cuvette less. It keeps the same dimensions so that they fit in the spectrophotometer cuvette compartment slots and keep the same path length (10 mm) so the incubation times are the same



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- 2) DO NOT DECOLORIZE. With the other kits, this is usually done with either PVPP or activated charcoal; however, these fining agents have been found to remove sulfite from the sample. Turbid samples may be filtered or centrifuged.
- 3) Standard needs to be prepared. In other kits from Vintessential, the standard already comes prepared in liquid solution. The kits are supplied with 2 ea. X 74 mg vials of lab grade sodium sulfite (≥ 99% purity – Winy or any other KMBS cannot be used for this). Each vial makes a 100 mL concentrated solution. That concentrated solution is then diluted with DI water to make the 50mg/L working standard (ratio = 1mL of concentrated solution to 9mL of DI water). The concentrate solution can be stored in the refrigerator for up to a week. The standards need to be prepared fresh and discarded after use.

Please note that the standard in this assay is used as a calibration factor (for calculation purposes only) and will not give a mg/L result. Expected A1 Standard absorbance is approximately 0.1, expected A2 Standard absorbance is approximately 1.2 - 1.4.

The indications supplied are based on our current knowledge and experience, but do not relieve the user from adopting the necessary safety precautions or from the responsibility of using the product(s) properly.

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Frequently Asked Questions

Q. I have heard that there can be some reliability issues with this type of analysis, such as not being able to get as accurate results with red wines vs. white, and other factors. Is that accurate?

A. These kits were designed based on the aeration-oxidation (AO) method. It is true that spectrophotometry uses light to go through a predetermined path length 10mm (or 1 cm) and it measures either the absorbance or the transmittance of light with a detector, depending on the assay. Because of this, color can cause an interference, and with most enzymatic kits there is sometimes a decolorization step required for highly colored red wines. In addition, because we want the reactions to happen in a reasonable amount of time (minutes not hours) there is typically a dilution step required. If the absorbance of the sample is too high, then you may need to dilute further or decolorize. However, for this test, the decolorization step is specifically not recommended because the act of fining color out with PVPP or charcoal etc., will remove some SO₂ resulting in false low measurements and, therefore, for this particular kit, only dilution is the acceptable way around the color interference.

Q. We currently have an AO set-up, which has its pros and cons, but is considered the "gold standard" in the industry (at least some might say that).

A. That is true, but again, these methods were developed based off AO and results compared against AO. If you look at the fact sheets provided you will see there is a 0.95 (R²) correlation for free SO₂ and 0.99 (R²) correlation for total SO₂ when comparing these manual spectrophotometer kits against AO.

Q. Why would you recommend going away from an AO set-up to this one?

A. With AO, one test takes 10 minutes just to aerate the sample. This does not include sample preparation, the titration, recording of the mLs used to titrate and a mathematical calculation. If you are really good, let's call it 13 minutes per sample. Of course, if you have two AO setups in tandem, you could run two samples in about 16 minutes and so on.

In addition, you are handling 25% phosphoric acid, which is dangerous. Hydrogen peroxide wants to constantly make itself turn into water, which means 3% H₂O₂ degrades quickly. 0.01 N NaOH is very diluted (this is 1/100th of 1N) and is constantly needing to be standardized in order to ensure the results are accurate. It is a lot of work and time consuming to do it right.

- Q. Considering sample prep, what would you consider "all in" time for running a sample?
- A. It really depends how skilled and experienced the operator is but anywhere from 2-10 samples in 30 minutes.

Free & Total SO₂ kits by Vintessential Laboratories for Discrete Analyzers:

Calibrate once weekly or as needed (for example, new reagents will need a new calibration). For this kit, users will need to buy sodium metabisulfite (98%) to generate your calibration curve. Keep in mind, your results will only be as good as how accurately you are able to make the standards.

Q. What is the accuracy range for these free SO₂ spectrophotometer tests?

A. The kits were designed and based off correlating the results with AO measurements. The correlation between test kits and AO: $R^2 = 0.95-0.97$ depending on the analyzer for free and total SO₂.

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For free SO₂, the calibrator range is 0–50 mg/L, so if free SO₂ is higher than this, this method may not be the best choice, or your own standards may need to be prepared and verified. The total SO₂ calibrator range is 0-250 mg/L in increments of 50 mg/L (ex: 0, 50, 100, etc.). Due to the volatility of SO₂, the standards need to be prepared, used and discarded after use. Fresh solutions should be prepared as needed. The instructions for preparing the calibrators are in the kit inserts.

Q. How long does it take to run a batch of 20 samples for F-SO₂ on a Chemwell-T?

A. Approximately 25-30 minutes. When running SO_2 samples, we recommend that samples do not sit exposed to air for extended periods of time before testing. Conveniently, the 2mL sample vials do have caps available. Simply sample your wine, cap and, when ready place in the instrument without delay.

Q. How many samples can I run in a batch?

A. For discrete analyzers (DA), 20 samples maximum at a time (Arena, Gallery, Kone, Chemwell T and Chemwell 2900 series). This is the maximum recommended because the more samples, the more time, resulting in possible false low results. You could probably run more but we do not have the data to support this. This applies to both free & total SO₂ for DA kits.

Q. How many tests can be run with one kit?

Α.

КІТ	TESTS PER KIT, GALLERY	TESTS PER KIT, ARENA/KONE	TESTS PER KIT, CHEMWELL-T	TESTS PER KIT, CHEMWELL (2910)
Free SO ₂ DA Kit	360	360	330	330
Total SO ₂ DA Kit	350	350	350	380

Q. Do I need to make my own calibrators?

A. Yes, and the customer needs to purchase their own sodium sulfite – it is not included in the kit like it does for manual spectrophotometer kits. It is recommended to use analytical grade sodium sulfite, and <u>instructions for the preparation of the calibrator standards</u> can be found on Vintessential's website.

Q. Do you provide the programmed methods for these kits?

A. <u>Gallery and Arena/Konelab</u>: The insert which is provided in the kit contains the method for users not using a Chemwell instrument (this is highlighted in the instructions). As with all other Vintessential kits, users can follow this method on the insert to program their Gallery and Arena/Konelab.

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<u>Chemwell:</u> The program methods in Chemwells are integrated into a new software update so anyone with a Chemwell will need to install this new software version which we can provide.

A new feature called **"true sample blanking"** has been added, which helps to eliminate any matrix effects from the sample. While running this method, users will notice that the unit uses TWO wells per sample, but they shouldn't worry. This is meant to happen and will provide them with the most accurate results.

As with any Chemwell software update, they must always download the software using Administrator Access. There is no need to uninstall the previous version - the new version should replace it automatically. They also should not need to save any of their own personal methods/data/etc. as they should reappear in the new software version if download is performed correctly. If there is anything that is particularly important, it should be saved on a computer for safe keeping. It is a good idea to review the software download section in the customer manual before doing this.

Q. Can I use these kits if I don't have a Chemwell, Thermo Arena, Gallery or Konelab?

A. There are many discrete analyzers on the market and it's impossible for Vintessential to design a method on each one. As long as the unit has the feature of true sample blanking like the Thermo and Chemwell units, then they should be able to get it to work. However, it may take some R&D on the volumes/std's/etc. on their behalf as all instruments handle the same chemistry differently.

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