



The use of inductively coupled plasma-atomic emission spectroscopy (ICP-AES) in the determination of lithium in cleaning validation swabs

Nancy Lewen*, Dennis Nugent

Bristol-Myers Squibb Co., Research and Development, Analytical R&D, 1 Squibb Drive, New Brunswick, NJ 08903, United States

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ABSTRACT

The pharmaceutical industry is required to perform cleaning validation studies to verify that equipment used in the manufacture of pharmaceuticals is adequately cleaned from one product or process to the next. Typically, these cleaning validation studies require an analytical method that uses some form of chromatographic technique. In the case of products that may have an inorganic constituent, however, it can often be easier to verify the cleanliness of equipment by using a non-chromatographic technique. A method is described to certify the cleanliness of processing equipment by determining lithium in cleaning validation swabs using inductively coupled plasma-atomic emission spectroscopy (ICP-AES).

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1. Introduction

Pharmaceutical companies are required to fully characterize their products as well as processes and raw materials that are used to make their products. Under cGMP regulations, the FDA requires that processing equipment be properly cleaned between production runs, and companies must be able to demonstrate that such equipment has been adequately cleaned [1,2]. Cleaning verification or validation methods are routinely developed to certify the cleanliness of equipment used, and because most active pharmaceutical ingredients (APIs) are organic compounds, these methods often involve some sort of chromatographic procedure, such as HPLC [3]. Other techniques, such as grazing-angle fiber-optic infrared reflection-absorption spectrometry (IRRAS) [4], desorption electrospray ionization (DESI) [5], total organic carbon (TOC) [6], and ion mobility spectrometry (IMS) [7] are also sometimes employed. Generally, these methods look for the presence of the active pharmaceutical ingredient (API) or components of the API either on the surfaces of the equipment that have been cleaned, and/or in the rinse solutions collected after the equipment has been cleaned [3,8].

Some pharmaceuticals are inorganic compounds or contain inorganic components. Cleaning verification of such substances can be accomplished by monitoring the concentration of a particular element associated with the compound under investigation, rather than the intact compound. This analytical strategy holds potential

advantages in terms of specificity, sensitivity, and sample throughput.

While the techniques of atomic spectroscopy have been used for the determination of lithium in biological matrices [9–13], and in the pharmaceutical industry for the purposes of monitoring for catalysts [14] and for the screening of pharmaceuticals for metals [15,16], these techniques have not normally been used for verifying the cleanliness of equipment in pharmaceutical processes. In the case of lithium carbonate, the active ingredient in many drugs for the treatment of bipolar disorder, using a technique more suited to the determination of lithium can be an easy and rapid means to demonstrate equipment cleanliness. Lithium carbonate is used in oral doses ranging from 500 to 2000 mg day⁻¹, with therapeutic effects seen in blood levels ranging from 2.8 to 8.3 mg Li l⁻¹ [17].

This work examines the use of inductively coupled plasma-atomic emission spectroscopy (ICP-AES) to evaluate the effectiveness of procedures to clean equipment after its use with lithium carbonate drug products. Although flame atomic absorption spectroscopy (FAAS) or graphite furnace atomic absorption spectroscopy (GFAAS) are more sensitive for the detection of lithium than ICP-AES, ICP-AES can be used for lithium concentrations >3 µg l⁻¹ [18]. Additionally, ICP-AES is a much less cumbersome technique than GFAAS, which after graphite tube conditioning, involves multiple steps (including drying, charring, atomization, and clean-out) that require optimization of temperatures, ramp and hold times for each of those steps, as well as sample volume and matrix modifier. As a result, ICP-AES provides a more rapid means of metals analysis than GFAAS, and it also provides greater flexibility than FAAS.

* Corresponding author. Tel.: +1 732 227 7534; fax: +1 732 227 3704.
E-mail address: Nancy.Lewen@bms.com (N. Lewen).

Inductively coupled plasma-mass spectrometry (ICP-MS) can also be used for the determination of lithium. However, with limits of detection of $<1 \text{ ng l}^{-1}$ [19], ICP-MS provides a means of analysis which far exceeds what is required for the determination of cleanliness of equipment used in the manufacture of lithium carbonate. Additionally, ICP-AES does not require the use of internal standards. With the addition of plasma spectrochemistry techniques in the United States Pharmacopeia (USP) [20], ICP-AES has become an accepted option for metals analysis in the pharmaceutical industry, and provides a good compromise between sensitivity, speed, and ease of analysis.

The method described uses filter flags, rather than the normally used cotton swabs, to collect the samples for analysis. Pieces of filter paper shaped like a flag, filter flags allow the swabbing of a surface with the rectangular “flag” portion, while holding the flag “pole.” The use of filter flags vs. cotton swabs provides for digestion of the complete swabbing material, with filter paper being much easier to digest than cotton. The method described has a limit of quantitation (LOQ) of $5 \mu\text{g Li}$ filter per flag, with the flags swabbing a surface area of 64.5 cm^2 ($0.08 \mu\text{g cm}^{-2}$).

2. Experimental

2.1. Reagents and materials

Concentrated nitric acid (Ultrex II grade) was purchased from VWR Scientific (West Chester, PA 19380). Type I water ($18 \text{ m}\Omega \text{ cm}$) was prepared by passing deionized water through a Barnstead water system (Thermo Scientific, Waltham, MA 02454). A certified NIST-traceable ICP stock standard solution containing Li at a concentration of $1000 \mu\text{g ml}^{-1}$ was purchased from Inorganic Ventures, Inc. (Lakewood, NJ 08701). Filter flags made of Whatman® #42 filter paper were purchased from Whatman® (Florham Park, NJ 07932). Acrodisc® filters from Pall Corp. (East Hills, NY 11548) were used.

2.2. Solvent selection and digestion parameters

In developing a method to determine lithium on filter flags, it was necessary to develop a procedure which would provide adequate dissolution of the filter flag and would also be compatible with the ICP-AES. Given that the filter flags are not readily soluble in deionized water, it was necessary to dissolve them with acid. Additionally, ICP-AES is a technique that performs best when samples are dissolved in either deionized water or dilute nitric acid. For this reason, a nitric acid digestion procedure was selected.

To determine the amount of acid needed for adequate digestion of the Whatman® #42-based filter flags, samples of Whatman® #42 filter papers were digested for 90 min at 105°C with final nitric acid concentrations of 25, 50, 75, and 100% in deionized water. Undiluted nitric acid provided the most complete digestion of the filter material. To determine the proper time period for the digestion procedure, blank filters were digested for 30, 60, 120, 180, and 240 min using 10 ml of concentrated nitric acid, with the 2-h time point yielding adequate digestion of the filter material.

2.3. Preparation of standard solutions

Working standard solutions were prepared containing 0.5 and $1.0 \mu\text{g ml}^{-1}$ lithium by diluting the lithium ICP stock standard solution in nitric acid:deionized water (20:80).

2.4. Preparation of samples

Working samples were prepared by carefully dampening a filter flag by pipetting $100 \mu\text{l}$ of deionized water onto the filter flag.

Table 1
Instrumental parameters for the ICP-AES method.

Parameter	Setting
Plasma view	Axial
Incident power	1300 W
Nebulizer flow rate	0.51 min^{-1}
Plasma flow rate	151 min^{-1}
Auxiliary flow rate	0.21 min^{-1}
Sample flow rate	0.8 ml min^{-1}
Read delay time	30 s
Resolution	Normal
Replicates	3
Spectral profiling	No
Peak algorithm	Peak area
Lithium wavelength	670.784 nm
Points per peak	3
Calibration equation	Linear through zero

The water was allowed to thoroughly soak into the filter flag. The presence of dry spots is acceptable, but care was taken to avoid saturating the filter flag. The filter flag was wiped over a 64.5 cm^2 (10 in.^2) section of the surface being examined, using the narrow end of the filter flag as the handle, and using the wide end of the filter flag to perform the swabbing of the surface.

The filter flag was placed into a 50 ml Digitube™ and 10 ml of concentrated nitric acid were added to the tube containing the flag. The tube was loosely covered and placed in a Hot Block™ and digested by heating at 90°C for 2 h. The digested sample was allowed to cool completely and the Digitube™ was diluted to its 50 ml volume with deionized water. A $1.2 \mu\text{m}$ Acrodisc® filter was used to filter any remaining residue.

2.5. Preparation of spiked samples

Spiked working samples were made by first preparing standard solutions to use for spiking. A series of standards containing 0, 0.10, 0.25, 0.50, and $1.00 \mu\text{g Li ml}^{-1}$ was prepared by diluting the lithium ICP stock standard solution in nitric acid:deionized water (20:80). Five filter flags were then spiked with the different Li standards and digested according to the procedure listed above for working samples.

2.6. Instrumentation

An Environmental Express Hot Block™ metal digestion system was used to digest the filter flag samples. A Perkin-Elmer Optima™ 4300 DV ICP-OES was used in this study. Table 1 lists the specific operating parameters used. The instrument was calibrated using working standard solutions that contained 0.5 and $1.0 \mu\text{g ml}^{-1}$ lithium.

2.7. Experimental procedure

Following the calibration of the instrument, the lithium standard solutions were assayed as samples to determine linearity of the method.

A series of filter flags was spiked and digested to determine the accuracy of the digestion procedure. The filter flags were spiked by pipetting aqueous standards of Li to just wet the flags and so that their final concentrations were 0.00, 0.10, 0.25, 0.50, and $1.00 \mu\text{g Li}$ per flag each. These filter flags were then digested for 2 h at 90°C in 10 ml of concentrated nitric acid. After digestion, they were allowed to cool and were brought to a final volume of 50 ml with deionized water, filtered and assayed to study possible matrix effects, as standard additions, as well as digestion efficiency.

Because various types of surface materials may be swabbed in the production environment, the accuracy of the method to

Table 2
Linearity of lithium standards.

Prepared concentration of Li ($\mu\text{g ml}^{-1}$)	Measured concentration of Li ($\mu\text{g ml}^{-1}$)
0.10	0.115
0.25	0.269
0.50	0.526
1.00	0.985
Slope	0.965
Intercept	0.028
Correlation coefficient	0.9995

adequately swab these potential surfaces and recover lithium that could potentially remain on the surfaces after cleaning was also investigated. Recoveries from nylon, stainless steel, Plexiglas, Teflon® and polycarbonate surfaces were all studied. In each case, a 64.5 cm² plate (10 in.²) of the surface material of interest was treated with 0.2 $\mu\text{g ml}^{-1}$ and then the filter flag was wetted with 100 μl of deionized water. The filter flag was then used to wipe the entire surface. Following this, the filter flag was placed into the digestion vessel with 10 ml of concentration nitric acid. Each filter flag was digested for 2 h at 90 °C. Once cooled, the samples were diluted to a final volume of 50 ml with deionized water and filtered prior to analysis.

3. Results and discussion

3.1. Linearity

Linearity of the method was studied by analyzing standard solutions containing five different concentrations of lithium, ranging from 0 to 1.00 $\mu\text{g ml}^{-1}$. Table 2 summarizes the results of the linearity study and demonstrates acceptable linearity and recovery for lithium, with a correlation coefficient of 0.9995 and slope of 0.965 (equivalent to a recovery of 96.5%).

3.2. Accuracy

Accuracy of the method was studied by performing a standard additions study to evaluate the recoveries of lithium from a series of filter flags that were spiked with lithium in the range from 0 to 1.0 μg per flag. Table 3 summarizes the results of this study, which indicate acceptable recovery and linearity for the procedure, with a slope of 1.047 (equivalent to a recovery of 104.7%) and a correlation coefficient of 0.9998.

Recoveries from nylon, stainless steel, Plexiglas, Teflon®, and polycarbonate surfaces were all acceptable, ranging from 90.5% for polycarbonate to 103% for nylon. The results of this study are provided in Table 4.

3.3. Specificity

The lithium wavelength at 670.784 nm is specific for lithium, with few coincident lines, and none that are commonly found (Sm,

Table 3
Results of standard additions study for lithium.

Prepared concentration of Li ($\mu\text{g swab}^{-1}$)	Measured concentration of Li ($\mu\text{g swab}^{-1}$)
0.000	0.001
0.100	0.106
0.250	0.259
0.500	0.505
1.000	1.051
Slope	1.047
Intercept	-0.003
Correlation coefficient	0.9998

Table 4
Results of surface swabbing study.

Surface material	Li placed on surface ($\mu\text{g swab}^{-1}$)	Li recovered from surface ($\mu\text{g swab}^{-1}$)	Recovery (%)
Nylon	0.20	0.205	103
Stainless steel	0.20	0.197	98.5
Plexiglas	0.20	0.188	94.0
Teflon®	0.20	0.196	98.0
Polycarbonate	0.20	0.181	90.5

Table 5
Reproducibility of replicate digested swabs (filter flags).

Parameter ($n=6$)	Value obtained ($\mu\text{g swab}^{-1}$)
Mean	0.103
Standard deviation	0.004
RSD (%)	3.45

for example). Recoveries of standards and spiked samples indicate no matrix effects, as well as no interferences from the filter flags or the inert surfaces tested.

3.4. Sample and standard stability

Because this method determines total lithium, the analyte will neither decompose nor degrade. Additionally, samples and standards are prepared in a nitric acid solution. As a result, sample and standard stability are not of concern. This is a departure from most chromatography-based analyses, where analyte degradation or decomposition is of a concern.

3.5. Precision (robustness)

The reproducibility of the method was studied by preparing and analyzing spiked replicates, containing 0.1 μg Li per flag. The results of this study are provided in Table 5 and indicate acceptable precision for the method, with an RSD of 3.45%.

3.6. Range

The range of the ICP-AES method was established based on the results obtained from the studies to demonstrate accuracy, precision, and linearity. The data obtained permits the conclusion that the range of the current method spans lithium concentrations from 0.1 to 1.0 μg Li filter per flag.

3.7. Limit of quantitation (LOQ)

As internal specifications required a LOQ of 2 mg Li filter per flag it was not necessary to attempt to achieve the lowest LOQ possible for the method. While the actual data indicate a lower limit of quantitation is possible, the present study was concluded at a limit of 0.1 $\mu\text{g ml}^{-1}$, which corresponds to 5 μg Li per flag, a value well below the target of 2 mg Li per flag.

3.8. Analysis of samples

Ten batches of swab samples were analyzed for their lithium content. All of the samples contained <5 μg Li per flag indicating that the equipment has been successfully and adequately cleaned.

4. Conclusions

While many analytical methods used to verify the cleanliness of production equipment used in the manufacture of pharmaceuticals employ chromatographic techniques to detect potential analyte(s),

some analytes may be more easily monitored using atomic spectroscopy. Additionally, unlike the frequently used cotton swabs from which it may be necessary to extract the analyte prior to analysis, filter flags may be used to swab equipment surfaces. The lithium concentration on equipment may be easily, rapidly, and accurately determined by ICP-AES and by using filter flags instead of cotton swabs.

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