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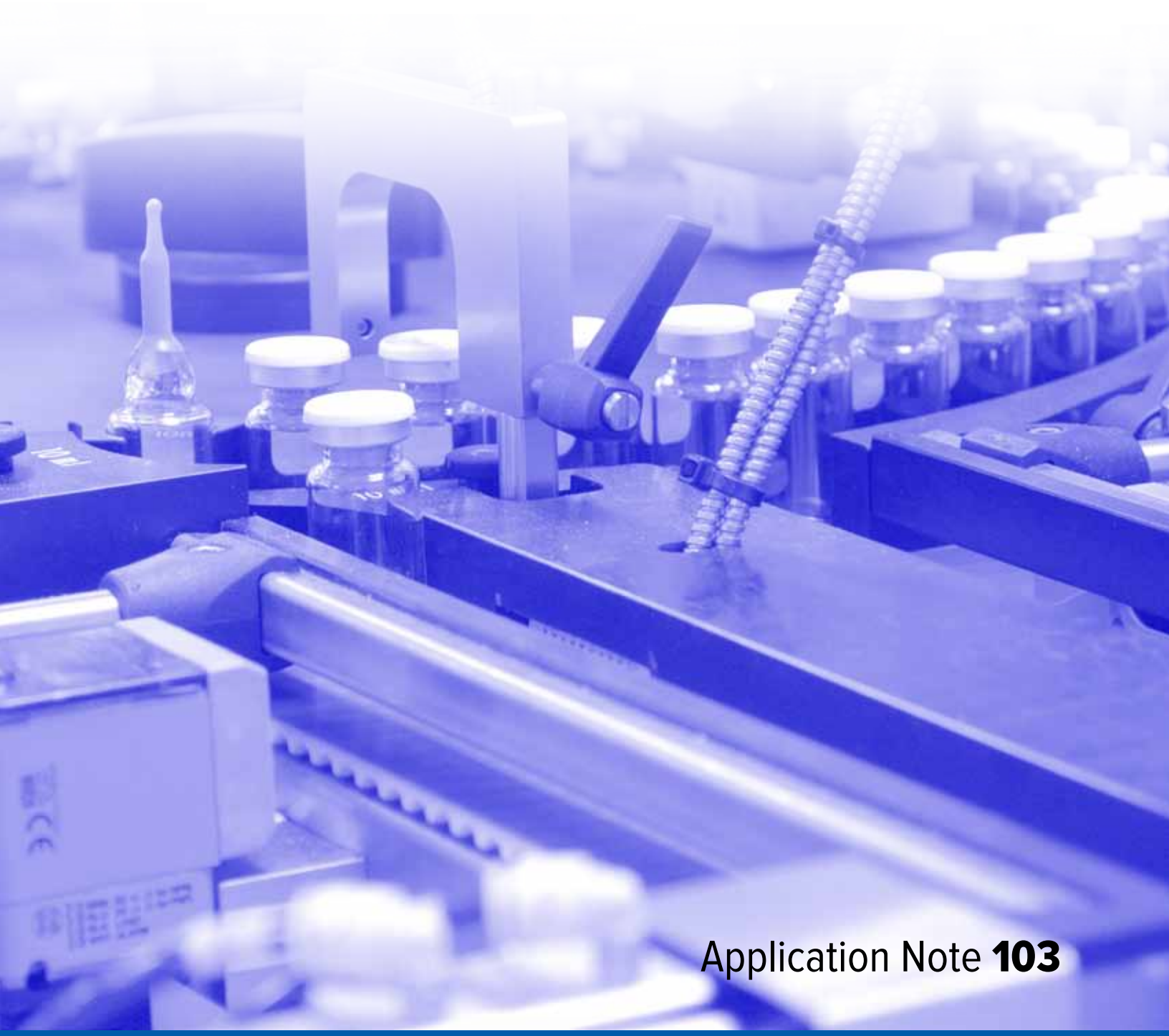
The Science of Pharmaceutical Manufacturing



Application Note **103**

Detecting Raised Stoppers in Sterile Freeze Dried Vials

Detecting Raised Stoppers in Sterile Freeze Dried Vials

Introduction

Lyophilization is a complex process that presents many manufacturing challenges, one of which is maintaining and monitoring container closure integrity of the finished package. Container closure integrity plays an important role in maintaining the sterility and stability of lyophilized products and concerns over patient safety, customer complaints, and the cost of investigations and product recalls are resulting in revised regulatory guidance (e.g. Annex 1 revisions to the European Guidelines for the Manufacture of Sterile Products). There is a corresponding drive in the industry to implement new types of inspection systems to help ensure the seal integrity of finished product vials. This includes vision sensor systems for 100% automatic detection of raised stoppers on vials containing lyophilized drug products prior to capping and sealing. A second type of inspection system which is increasingly being implemented is laser-based machines for 100% headspace inspection. This Application Note describes how laser-based headspace inspection detects vials having raised stopper issues. Data from 'raised stopper vials' is presented showing that quantifying the physical headspace conditions also enables science-based process trouble-shooting and optimization.

If the rubber stopper on a vial is not seated properly before entering the capper, the effects can be twofold. First, the stopper could be pulled off and product spilled, and second, the stopper could be pushed back into the vial and reseated. If the stopper is reseated on a vial that has been leaking, there is a risk that sterility could be compromised without detection (the Annex 1 revisions to the European Guidelines for the Manufacture of Sterile Products address this point by requiring the use of grade A sterile air to protect stoppered vials until capping). Many companies are implementing vision sensors that determine the stopper position before capping to detect any vials having raised stoppers. Typically, two cameras are used to ensure that the stopper is seated around the whole circumference of the

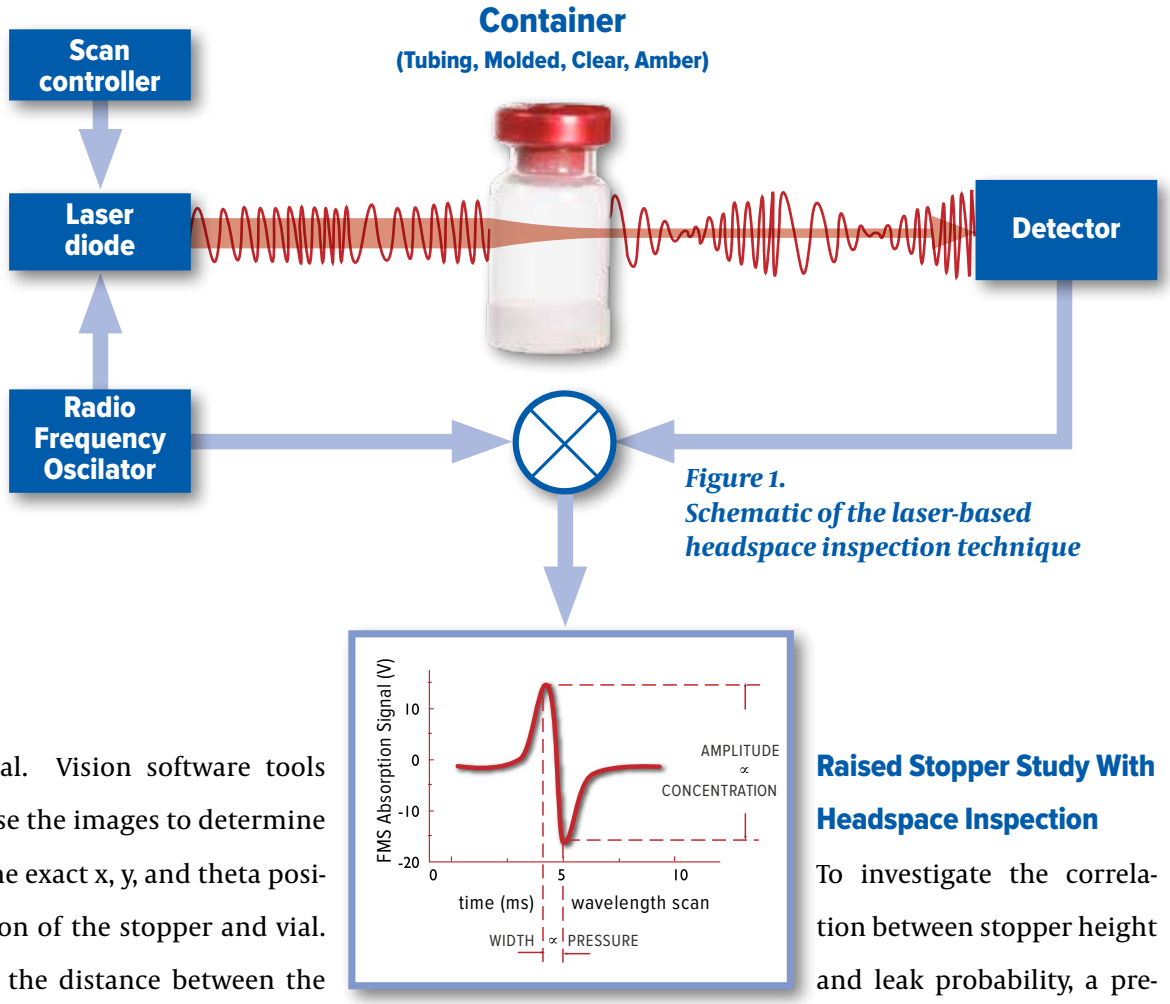


Figure 1.
Schematic of the laser-based headspace inspection technique

vial. Vision software tools use the images to determine the exact x, y, and theta position of the stopper and vial. If the distance between the top of the vial and the lip on the stopper exceeds a predetermined distance then the vial can be rejected. Although validating such a vision inspection method can be relatively costly, implemented systems have proven to accurately determine stopper position. The disadvantage of this type of inspection is that stopper height does not absolutely determine if a vial has leaked or not. Stopper height only correlates to a probability that a vial has leaked. Headspace inspection however does provide a direct determination of container closure integrity and absolute confirmation of whether a vial has leaked or not.

Raised Stopper Study With Headspace Inspection

To investigate the correlation between stopper height and leak probability, a preliminary study was carried out in a recent collaboration between Aptuit and LIGHTHOUSE. The laser-based headspace inspection platforms used in the study utilize a high sensitivity detection technique known as Frequency Modulation Spectroscopy (FMS). Light from a near-infrared semiconductor laser is tuned to match an internal vibrational frequency of either the oxygen or water molecule. Measuring the absorption of the laser light after it passes through the container headspace allows for the determination of headspace pressure, oxygen, and moisture concentrations. Figure 1 displays a schematic of the FMS technique. To

simulate vials having different raised stopper heights, plastic shims were cut into a U-shape with "legs" 20 – 25 mm long. The space between the legs was such that the U-shaped shim was a neat fit around the stopper stem just under the stopper flange. The base of the shim U was positioned immediately above the single igloo door of the stopper. Shims of 0.5, 1.0 1.5 and 2.0 mm thickness were placed on 40 stoppers. The stoppers were then partially inserted into 5ml vials and the vials put into a Lyostar I development freeze dryer. The freeze dryer was evacuated to <0.05 mbar. The chamber was then back-filled with nitrogen to 400 mbar and the vials stoppered in situ. The chamber was then back filled to 1000 mbar with nitrogen, the chamber door opened and the vials removed.

The vials were left uncapped under ambient air conditions and after one hour the shims were withdrawn, using pliers, while maintaining by hand a downward pressure on the stopper to avoid accidental displacement. The vials were then over-sealed with aluminum caps after the one hour wait time.

Each vial was then measured for headspace oxygen and pressure using the non-destructive

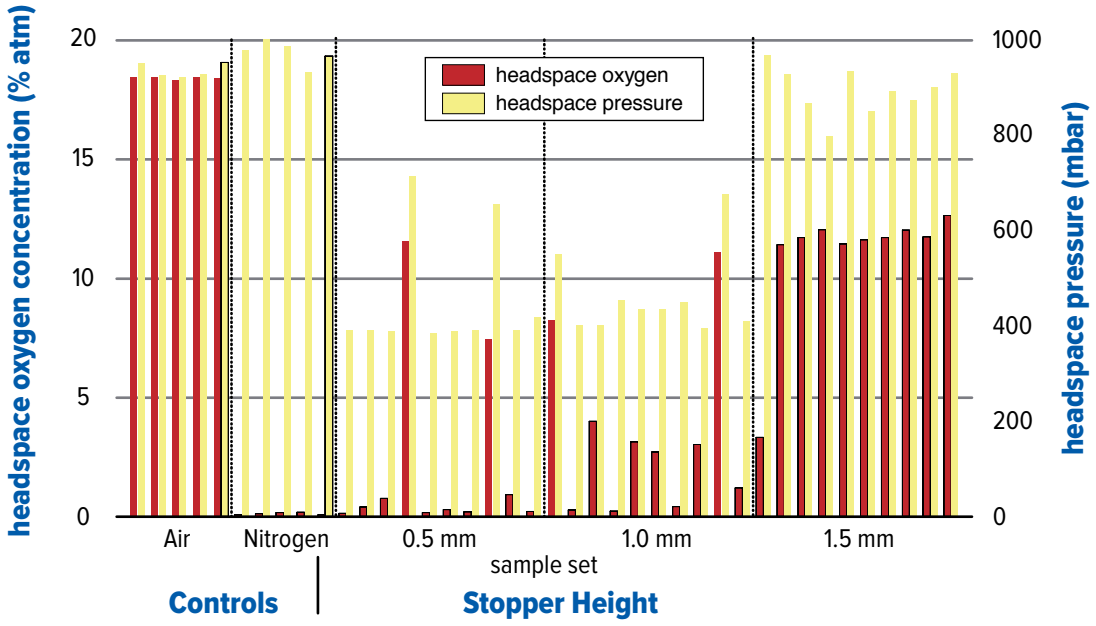


Figure 2.
Headspace oxygen and headspace pressure data from the raised stopper experiment. Laser-based headspace inspection reveals that a leak is a probabilistic function of the raised stopper height. Also plotted are sets of air and nitrogen control vials.

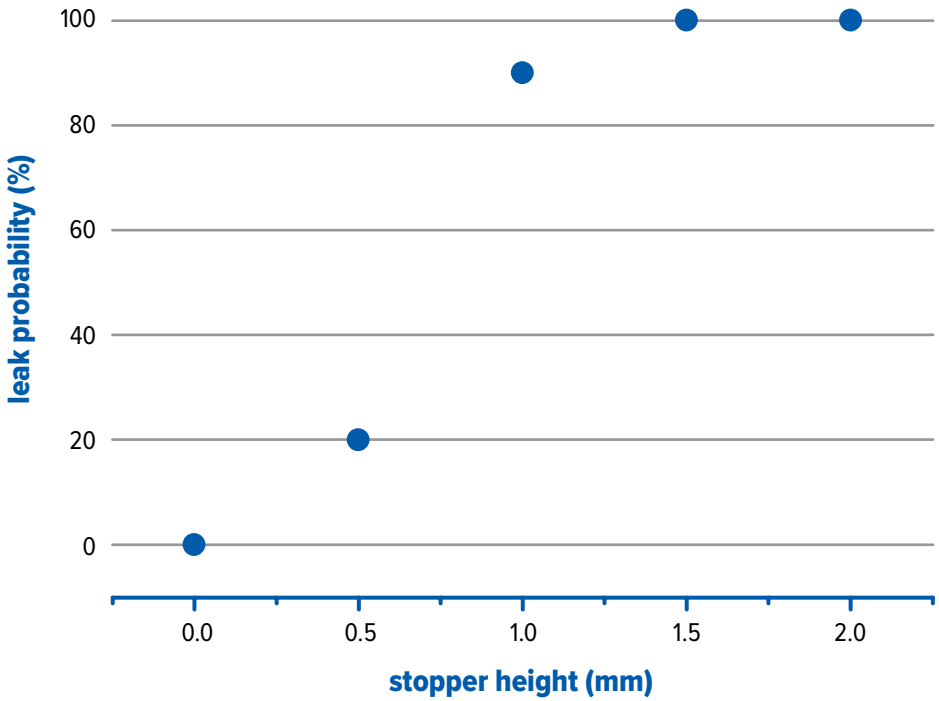


Figure 3.
Plot of leak probability as a function of stopper height.

laser-based headspace inspection platforms from LIGHTHOUSE. If the oxygen level was above 1% or the pressure was above 420 mbar then the vial was considered to have leaked. The results of the headspace measurements are shown in Figure 2.

The data in Figure 3 show the percentage of vials that leaked as a function of the stopper height. For the vials with no shims 0 of 10 vials leaked. For the vials with 0.5mm shims, 2 of 10 vials leaked. For the vials with 1.0mm shims, 9 of 10 leaked. For the vials with 1.5mm shims, 10 of 10 leaked. For the vials with 2.0mm shims, 10 of 10 leaked.

Two conclusions can be drawn from these data. First, the closure failure rate is a probabilistic function of the stopper height. Simply rejecting vials based on the stopper height means some good vials will be rejected and some bad vials will be accepted. Second, headspace inspection provides a direct measure of container closure integrity by detecting physical changes in the headspace conditions of a leaking vial. Implementation of headspace inspection after capping enables the detection of all

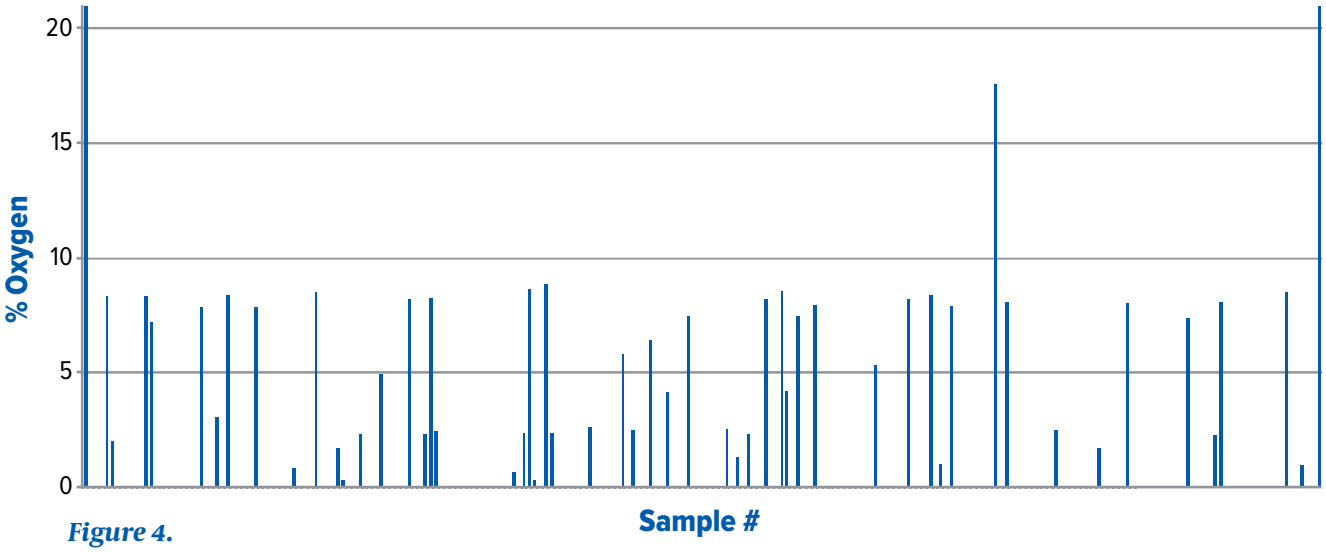


Figure 4.
Plot of headspace oxygen levels showing that a high percentage of product vials have leaked ingressing air into the headspace.

vials that have leaked, even when the leak was transient to some point in the manufacturing process. It should also be noted that headspace inspection detects not only leaks due to raised stoppers, but also leaks due to cracks in the glass vial, defects under the aluminum seal, out-of-specification vials/stoppers, etc.

Industry Case Study

This section describes an industry raised stopper case study involving a commercial batch of lyophilized product. The batch consisted of approximately 11,000 vials stoppered at 600 mbar of nitrogen. A suspected raised stopper issue motivated the manufacturer to perform 100% laser-based headspace inspection of the batch a few weeks after production. To identify any potential leaking vials and to gain insight into any potential process issues, Total Headspace Characterization (THC™) was performed meaning that product vials were inspected for headspace oxygen, headspace pressure, and headspace moisture levels.

Figure 4 shows the headspace oxygen results for a subset of the product samples. It is clear from these results that a significant percentage (~ 25%) of the product vials have high oxy-

gen content, presumably due to leaks that ingress air into the headspace. It is interesting to note that a large proportion of the high oxygen product samples have oxygen levels around 8% and that only one product sample contains near atmospheric oxygen levels.

The measured headspace pressures give additional insight into the quality of the batch and are plotted in Figure 5. In this particular graph, the headspace pressures of the high oxygen content vials are plotted towards the end (righthand side of the graph). It is now clear that the high oxygen content vials have lost vacuum relative to the specified stoppering pressure of 600 mbar with a large proportion of the defective vials showing headspace pressures near one atmosphere.

The correlation between the headspace oxygen and headspace pressure measurements in he defective vials is plotted in Figure 6 and provides the deepest insight into the closure integrity issue of this particular batch. Analysis of these data identifies three categories of leaking vials:

Category 1 – Temporary gross leaker: From Figure 6 it can be seen that the vials having 8% headspace oxygen levels also have headspace pressures near one atmosphere. These vials suffered from significantly raised stoppers and were grossly leaking during the few minutes from exiting the freeze dryer to being transported to the capping machine. Due to the

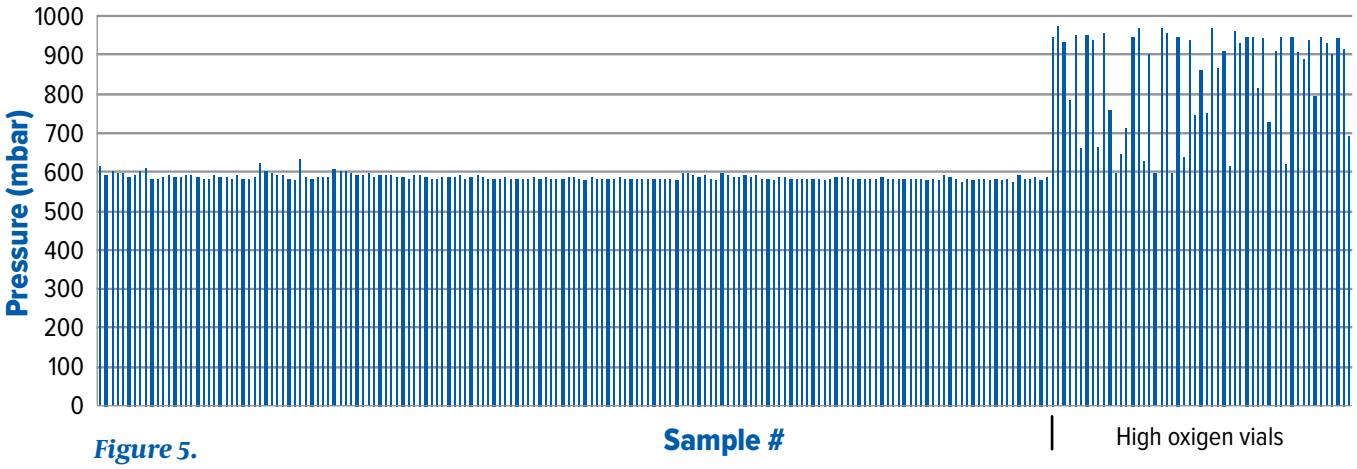


Figure 5.
Plot of headspace pressure levels in the product vials. Most vials are at the specified stoppering pressure of 600 mbar. The pressure of the high oxygen vials are plotted on the righthand side and confirm that these vials have suffered from partial or full vacuum loss.

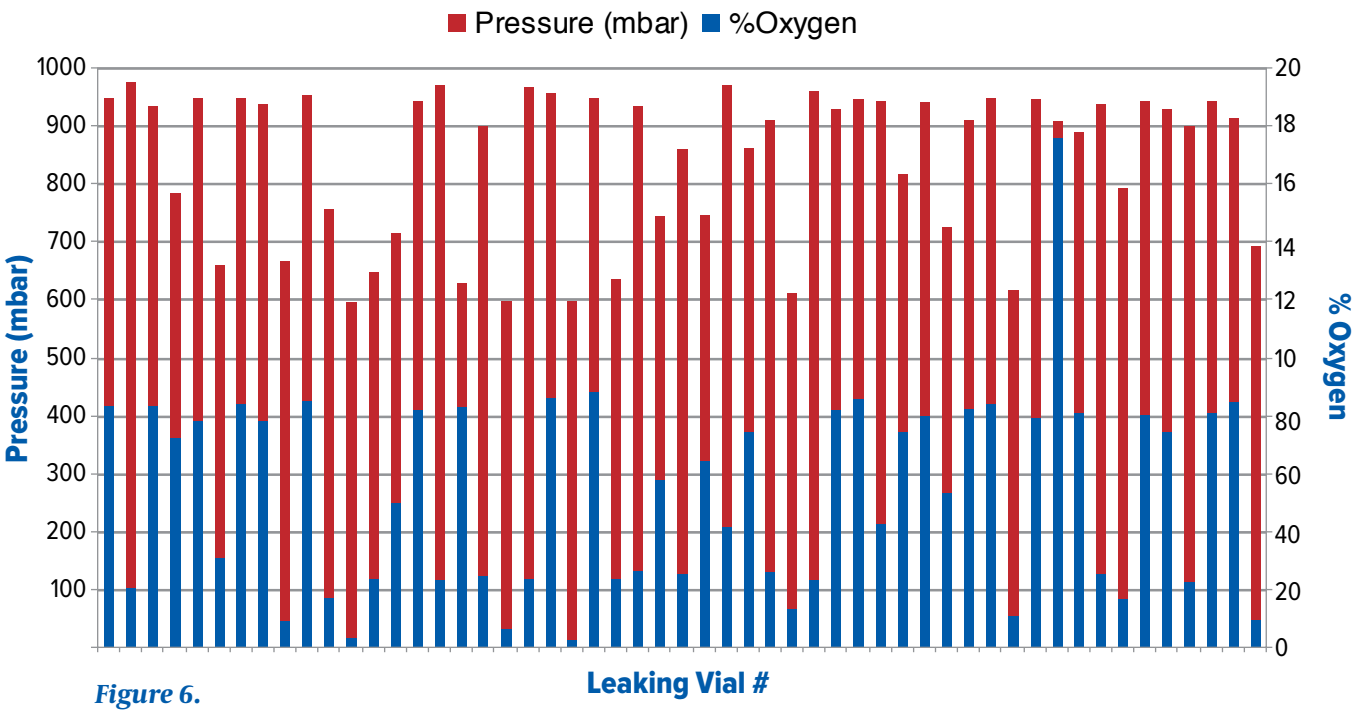


Figure 6.
Plot showing the correlation between headspace oxygen and headspace pressure in the leaking product vials.

gross leaks, 400 mbar of air (corresponding to 8% oxygen) quickly ingressed into the product vials. The capping process stopped the leaks sealing these vials under headspace conditions of 8% oxygen and one atmosphere.

Category 2 – Permanent leaker: One vial in Figure 6 can be seen to have near atmospheric oxygen and pressure levels. It can be concluded that this vial also had a raised stopper coming out of the freeze dryer resulting in a leak. In contrast to the vials described in Category 1, the capping process did NOT stop the leak in this vial. After an initial ingress of 400 mbar of air into the headspace driven by the pressure differential inside and outside of the vial, this vial continued to ingress air after capping through a diffusion process resulting in headspace gas exchange. In this case, the headspace oxygen content continued to rise until it matched atmospheric oxygen levels.

Category 3 – Temporary partial leaker: The remaining defective vials in Figure 6 have oxygen levels between 0.5% up to 8% and headspace pressures between 600 mbar up to one atmosphere. It can be concluded that these vials had slightly raised stoppers resulting in

smaller leaks. In the few minutes between the freeze dryer and the capping machine, these vials ingressed some amount of air partially raising the headspace pressures above 600 mbar. The leaks in these vials were stopped by the capping process sealing the vials under a range of partially elevated headspace oxygen and pressure levels.

Finally, Figure 7 shows the results of the headspace moisture inspection performed on the product vials. Again, the results for the defective vials are plotted on the righthand side of the graph. Elevated water vapor levels in the headspace can also be a sign that the vial has leaked. A leaking vial ingresses humid air resulting in increased headspace moisture. Vials that are not leaking can also have elevated headspace moisture levels. In this case, the elevated moisture points to an issue with the drying process and can identify vials that have not been dried to specification. The average headspace moisture content of the defective vials in Figure 7 is approximately 20% higher than the average moisture content of the good

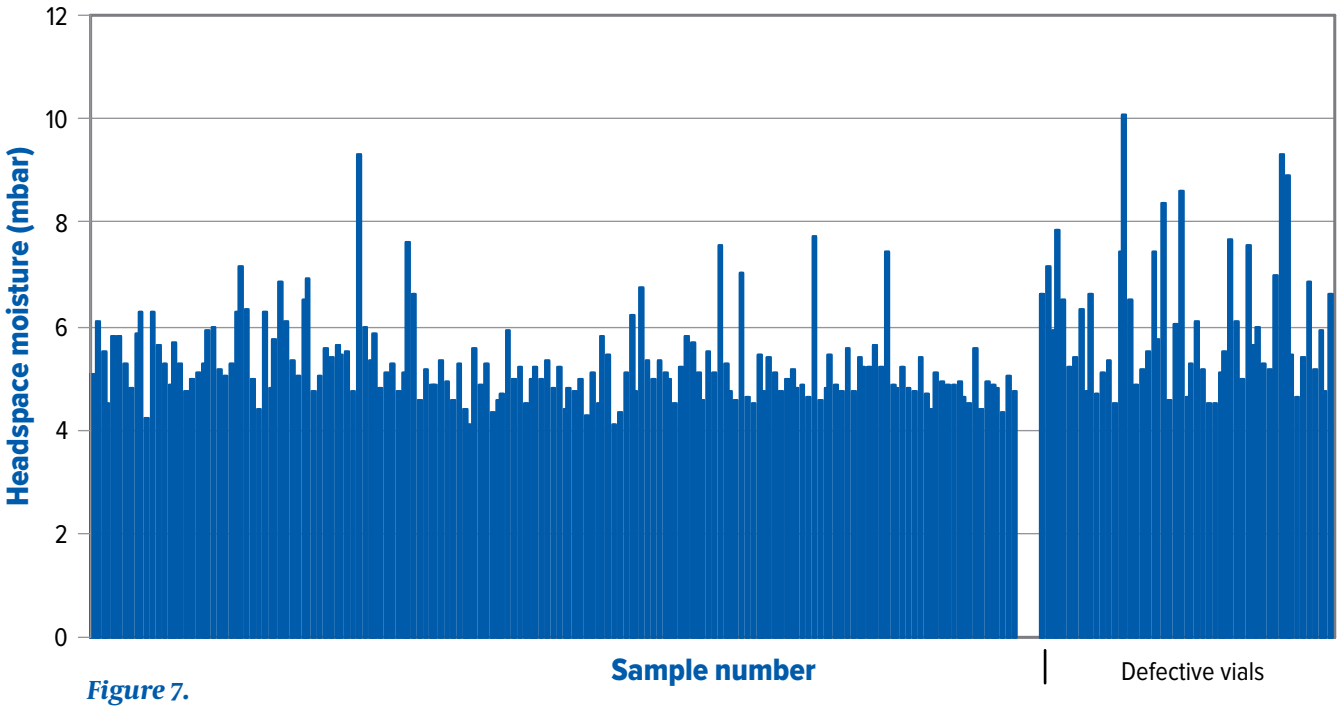


Figure 7.
Plot of the headspace moisture levels in the product vials. The headspace moisture levels of the leaking vials are plotted on the righthand side.

vials. This confirms that the defective vials did indeed ingress some amount of humid air into the headspace in the time between exiting the freeze dryer and being capped. It is interesting to note that even in the well sealed vials there are vials that display elevated headspace moisture levels. This indicates that the freeze drying cycle could potentially be optimized for more homogenous drying across the batch.

Conclusions

The studies described in this Application Note give insight into the detection of raised stoppers and the corresponding loss of container closure integrity. The use of vision sensors can be used to determine stopper height, but correlating stopper height to a leak in a sterile vial can be problematic. The results presented here show that closure failure rate is a probabilistic function of the stopper height. Setting a non-zero stopper height as a reject limit before capping will result in the rejection of good vials and, more problematic, the acceptance of bad vials.

On the other hand, laser-based headspace inspection is a direct measure of container closure. The physical headspace conditions of a freeze-dried vial will change due to a leak. Quantifying the headspace conditions provides a wealth of information in addition to the identification of vials that have leaked. The industry case study presented here shows how Total Headspace Characterization (THC™) identifies vials that have lost closure and gives specific insight into where in the manufacturing process the problem has occurred. Although not discussed in this Application Note, Total Headspace Characterization can also be used to identify vials posing a stability risk due to elevated moisture or oxygen levels. Current industry implementations of Total Headspace Inspection machines are part of a robust finished product inspection process. Headspace inspection machines implemented in packaging departments are performing 100% container closure integrity inspection and guaranteeing that any leaking vial is identified and removed from the line. In addition to providing science-based insight into the process, these headspace inspection implementations address the current regulatory guidelines to guarantee container closure integrity and the maintenance of sterility (and stability) of finished lyo product.

About Us

LIGHTHOUSE is the leading manufacturer and provider of laser-based headspace inspection systems for finished sterile product applications specific to the pharmaceutical industry. These applications include leak detection, container closure studies, headspace oxygen monitoring, product moisture determination, and lyo cycle development in both production and R&D environments. LIGHTHOUSE developed the laser-based headspace technology with funding from the Food and Drug Administration. We have hundreds of headspace inspection systems installed around the world at some of the world's leading pharmaceutical, biopharmaceutical and contracting manufacturing companies including: Amgen, Baxter, Bayer, Boehringer Ingelheim, BMS, Covidien, DSM, Eli Lilly, Genentech, GlaxoSmithKline, Helvoet Pharma, Johnson & Johnson, Merck, Novartis, Patheon, Pfizer, Roche, Schering-Plough, Serum Institute of India, Sankyo, Sanofi-Aventis, Talecris Biotherapeutics, TEVA, West Pharmaceutical Services, and Wyeth.

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