



Monitoring Airborne Micro-organisms in Blow-Fill-Seal Technology

Article Reprinted from the ©June 2004 issue of:

**Pharmaceutical
Technology**
EUROPE

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Monitoring Airborne Micro-organisms in Blow-Fill-Seal Technology

The article describes the basic principles of blow-fill-seal (BFS) technology together with the advantages it offers. Although BFS technology is an ideal process for aseptic filling of liquid pharmaceutical products there is still a risk of contaminating the product inside the filling area. This, together with regulatory requirements for the microbiological control of critical areas in pharmaceutical production, makes microbiological monitoring a necessity. Two concepts will be presented in this article, describing how microbiological monitoring of BFS machines can be carried out in the critical filling zone during production without any risk of contamination and complying with the relevant guidelines.

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Blow-fill-seal (BFS) technology is being increasingly used for aseptic liquid filling. The European Union (EU) Guide for Good Manufacturing Practice (GMP) (revision of Annex 1, Manufacture of Sterile Medical Products) defines BFS machines as “... units that are purpose-built machines in which, in one continuous operation, containers are formed from a thermoplastic granulate, filled and then sealed, all by one automatic machine.”¹ Thus, BFS machines manufacture the containers to be filled, fill them and then seal them immediately, with all steps performed rapidly and within a small space.² The individual stages of the BFS process are illustrated in Figure 1.

BFS technology allows containers to be unit doses, multidose ampoules, vials or bottles. Compared with

conventional methods, the technology has the advantages of

- preservative-free products (unit doses)
- minimized time and space
- automation
- increased product safety (microbial-free).

Despite these advantages, BFS technology always entails a potential risk of contaminating the product within its small space. For this reason, the liquid to be filled and the container are protected from any possible contamination during the filling process by a stream of filtered sterile air. The relevant guidelines¹ are increasingly calling for testing the microbiological quality of BFS machines. EU guidelines classify the filling area of a BFS machine as a grade A environment; that is, the concentration of micro-organisms

must be <1 cfu/m³ air (where cfu is colony-forming units). Figure 2 shows a typical set-up for microbiological monitoring of BFS machines, such as that used at Holopack (Abtsgmünd-Untergröningen, Germany).

The measuring system

Monitoring air directly at the point of filling means that a few obstacles must be overcome: limited space, poor accessibility, movable parts and a latent risk of contamination. This makes it necessary to withdraw the air sample from the interior of the BFS machine whilst the air sampler remains outside. The monitoring system (as used in Figure 2) consists of an air sampler (pump and flow rate meter), a reusable metal filter holder connected to the air sampler by a hose and an easy-to-exchange disposable gelatin filter.

The hose connection enables sampling to take place within the filling area whilst the air sampler itself remains outside the critical area. During sampling, the air sampler draws a defined volume of air through the gelatin filter; after sampling, the filter is placed on a suitable culture medium in a petri dish and incubated. The colonies formed are counted and evaluated as the number of cfu/m³ of air.

Water-soluble gelatin filters have a pore size of $3\ \mu\text{m}$. They are sterilized by gamma irradiation and are capable of retaining 99.9995% of *Bacillus subtilis niger* spores (at an air flow rate of $0.25\ \text{m/s}$)³ and 99.94% of coliphages T3 (at 80% relative humidity [RH]).⁴ The air sampler can be calibrated on site and has been optimized for use in critical areas such as grades A and B (class 100) clean rooms, isolators and BFS machines. Two different concepts for air monitoring in BFS machines are introduced below.

Concept 1: measurement with the hose between sampling point and filter holder. This concept describes the example of Excelvision, a pharmaceutical manufacturer based in Annonay (France). At the company's plant, BFS machines (*Bottlepack 3012*; Rommelag, Switzerland) were monitored. The machine is used to fill plastic vials of capacities ranging from 0.3–5 mL with eye drops.

Figure 1 The stages involved in the BFS process (courtesy of Rommelag AG).

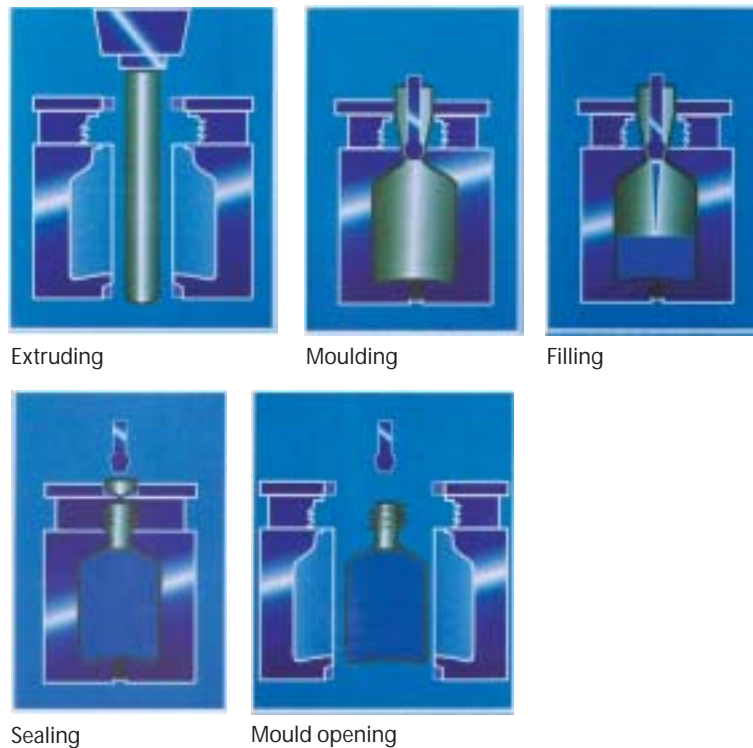


Figure 2 Bottlepack machine (located at Holopack) with the filter holder attached near the filling zone and with the MD8 airscan.



Sampling was performed using a polyvinylidene fluoride (PVDF) hose of sufficient length (Figure 3) that was connected directly to the filling area (classified as grade A). The hose was routed approximately 2 m

to the outside, where it was attached to a metal filter holder specifically designed for use with disposable gelatin filters (Figure 3 inset). The hose was attached to the disposable gelatin filter (*17528-80-ACD*) by a bayonet catch. To prevent secondary contamination, the filters were changed inside a *Captair Flowcap700 S* clean bench, the interior of which was also classified as grade A. Through a further connected hose, the air drawn from the clean bench was pumped through the air sampler into the grade C clean room environment, where the air sampler was located.

To monitor the filling area of the BFS machines, one air sample per machine per week was taken at an air flow rate of $6\ \text{m}^3/\text{h}$ for 10 min. This corresponded to a total sampled air volume of $1\ \text{m}^3$ in 10 min, which meets the requirements of EU GMP regulations. The culture media (tryptic soya agar [TSA]) were incubated for 3–5 days at $32.5 \pm 2.5\ ^\circ\text{C}$. The results showed that the values were less than the limits of <1 cfu/m³ required for grade A clean rooms. For example, no microbes ($0\ \text{cfu/m}^3$ of air) were detected from 29 June 2000

Figure 3 Bottlepack sampling (located at Excelvision) using the PVDF hose, which is connected directly to the filling area.



to 13 February 2001 for 31 batches. The standard operating procedure (SOP) for air monitoring has been in place at Excelvision for 3 years and no contamination has ever been detected.

Concept 2: measurement using a metal filter holder containing a gelatin filter and connected directly at the filling zone. Another possibility for air sampling in BFS machines is using the *MD8 airscan* with a specially manufactured, reusable metal filter holder (Figure 4) in which a disposable gelatin filter (*17528-80-ACD*) is inserted. For sampling, the gelatin filter disposable in the filter holder is located in the direct vicinity of the filling zone. This concept is based on the example at Holopack, which uses a *Bottlepack 3012M* to fill sterile products (small volume parenterals, water for injection and isotonic physiological saline) in plastic vials with capacities ranging from 10–40 mL. In this measurement set-up, the filter holder with the disposable gelatin filter is located near the filling zone (grade A), in which the filling needle is located (Figure 2). The specially fabricated metal filter holder, which has to be sterilized before each sampling procedure, is connected to the BFS machine by a special sanitary fitting. In this example, air sampling was performed once per lot (after filling) at an air flow rate of 6 m³/h for 10 min

Figure 4 The special metal filter holder (located at Holopack).



per sample. Thus, 1 m³ of air was sampled in 10 min per lot. The culture medium (CASO) was incubated at 30–35 °C for 5 days. The results showed that during 2003, from week 3 to week 25, no contamination was found for a total of 90 lots (that is, 0 cfu/m³ air). Hence, the filling zones of the BFS machines complied with EU GMP requirements (<1 cfu/m³ room air in grade A environments).

Validation

As part of validating the two methods, it must be ensured that 1 m³ of air per procedure must be sampled; therefore, it is necessary to calibrate the air sampler. Additionally, it must be ensured that the risk of secondary contamination is eliminated during sampling to prevent false positive results. As shown here, this can be done by using a clean room grade A clean bench when changing the disposable gelatin filter, among other measures. Further, any microbes present in the filling

zone must not be allowed to escape detection through impaction within the hose, which could lead to false negative results. This means that for sampling according to concept 1, the length, inner diameter and the roughness of the hose must be optimized and validated. If concept 2 is applied, validation of the hose is not necessary as the filter holder with the gelatin filter is connected directly to the filling zone.

Summary

Today, the gelatin membrane filter method is a recognized procedure for air sampling in critical clean room environments and isolators. As described in this article, it is also ideal for monitoring air in the critical filling zone of BFS machines because it can be used during production without entailing any additional risk of contamination and it complies with the relevant guidelines. In the future, it is expected that BFS technology will continue to gain significance similar to isolator technology. With its advantages, the gelatin membrane filter method will be increasingly used for testing BFS systems, as is already the case for isolators.

Acknowledgements

I would like to thank Benoit Carles, microbiology control head at Excelvision, and Walter Matheis, QC manager at Holopack Verpackungstechnik GmbH for providing images and data.

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The original German-language version of this article was first published in the journal *Steriltechnik*, 3rd issue, no. 2, Sept. 2003 (3. Jahrgang, Nr. 2). ■