

OPERATING MANUAL



VIABLE (MICROBIAL) PARTICLE SIZING INSTRUMENTS

**Six Stage and Single Stage
Models BGI10800 Series & BGI10890**



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**Viability (Microbial) Cascade Impactors
Six Stage Impactor BGI Model BGI10800**

INTRODUCTION

The study of the microbial content of the air has been of recognized importance for over a century. Significant developments during WWII placed an even sharper emphasis on this scientific area. Since the events of September 11, 2001 Aerobiology has become an important component of Air Pollution. Cascade impaction is and remains the premier method of accurately quantifying the viable content of an atmosphere. Biological aerosols are defined as viable biological contaminants occurring as solid or liquid particles in the air. These particles can vary in size from viruses less than 0.1 micrometer in diameter to fungal spores 100 micrometers in diameter. They may occur as single, unattached organisms or as aggregates.

Viable particle samplers have been used to collect and study aerobic species of bacteria and fungi. Even though many viable samplers, including the BGI Instruments Sampler, will collect some virus particles, there is no convenient, practical method for the cultivation and enumeration of these particles. There are two constraints on all viable particle samplers. First, the particle must be separated from the air for any viability study, and second, the ability to reproduce (viability) must be demonstrated. Sampling and microbiological lab techniques are combined. The purpose of this manual is to outline proper methods for the study of biological aerosols using BGI Instruments (BGI) Viable Particle Samplers.

Six-Stage Viable Particle Cascade Impactor

The BGI 1CFM (28.3 liter per Minute) at the Actual Temperature and Barometric Pressure six-stage Viable Particle Sampler is a multi-orifice cascade impactor which is normally used to measure the concentration and particle size distribution of aerobic bacteria and fungi in the ambient air. The instrument has been widely used as a standard for enumerating the viable particles in a microbial aerosol. Viable particles can be collected on a variety of bacteriological agar and incubated in situ for counting and identification. Typical nutrient agar is a common bacteria growth media.

This sampler is flow calibrated in such a manner so that all particles collected, regardless of physical size, shape or density, are sized aerodynamically and can be directly related to human lung deposition. The viable samplers may be briefly described as follows:

- a) Collection plates are prepared by aseptically pipetting 27ml of sterile, bacteriological agar (45-50° c), into each of six glass Petri dishes. Petri dishes other than those supplied, cannot be used since this would alter the distance between the jet orifice and the collection surface of each stage. Plastic Petri dishes should not be used because the static charge generated reduces the collection efficiency of the process.

- b) Any, general purpose, solid bacteriological medium, such as trypticase soy agar, or blood agar can be used for the collection plates. Selective media are not recommended since they inhibit the repair and growth of injured or stressed cells.
- c) One collection plate, with the cover removed, is inserted on each stage of the sampling instrument.
- d) The air to be sampled enters the inlet cone and cascades through the succeeding orifice stages from Stage 1 to Stage 6. Due to successively higher orifice velocities, smaller particles are inertially impacted onto the agar collection surfaces.
- e) Viable particles are retained on the agar plates, and the exhaust air is carried through the vacuum hose to the vacuum source (individual pump or in-house vacuum system).
- f) For maximum collection efficiency, a constant flow is necessary. This may be provided with a continuous-duty vacuum pump. Periodic calibration is recommended (See Calibration Section). Another method of assuring a constant flow would be to insert an airflow meter (not provided), with a minimum capacity of 1 ACFM (28.3 liters/min.) in the vacuum hose between the sampler and the vacuum source. The user should calibrate this flow meter, using a Rotameter or electronic flow meter of the non pulsating type.
- g) After sampling has been completed, the sampling time is recorded and the agar collection plates are removed from the sampling instrument. The cover is replaced, on each Petri dish. Identify each plate as to sample and stage number A (i.e., 1-1, 1-2, 1-3, etc.).
- h) Place all agar plates, inverted to prevent condensation drip, in an incubator at 35°C for 18 to 24 hours. Plates can be incubated at room temperature if the user is most interested in environmental bacteria whose optimum growth temperature is lower than body temperature or at 20° to 25°C for maximum recovery of fungi.
- i) After incubation, the number of colonies on each plate is counted, using a standard bacterial colony counter.
- j) Knowing the air sample flow rate and the sampling time, the mean number of viable particles (aerobic bacteria and/or fungi) per unit volume of air can be calculated, and the percent or particles in each size range can be estimated.

Aerodynamic Particle Sizing

The design of the BGI Viable sampler was developed from the following reality:

The human respiratory tract is an aerodynamic classifying system for airborne particles. A sampling device can be used as a substitute for the respiratory tract to act as a collector of viable airborne particles. As such, it should reproduce, to a reasonable degree, the penetration of the lung by these particles. The fraction of inhaled particles retained in the respiratory system and the site of deposition vary with all the physical properties (size, shape, density) of the particles

which make up the aerodynamic dimensions (Figure 1). The pulmonary deposition of unit density particles is known. The particle sizes that are collected on each stage of the BGI Viable Samplers have been determined. Therefore, if a standard model of these samplers is used according to a standard operating protocol, the stage distribution of the collected material is comparable to deposition in the various regions of the lung. Figure 1 shows the deposition efficiencies in the nasal-pharyngeal, tracheo-bronchial and pulmonary regions of the human respiratory tract as a function of particle size. Large particles deposit primarily in the nasal-pharyngeal area, whereas particles of sub-micrometer size deposit, mainly, in the pulmonary area.

Numerous small round jets improve collection (impaction) efficiency and provide a sharper cutoff of particle sizes, on each stage of inertial impactors. Thus, the Six-Stage sampler, with 400 small round jets per stage, meets all the criteria for the efficient collection of airborne viable particles. Reports have discussed a reduced efficiency in cascade impactors when particles bounce off the

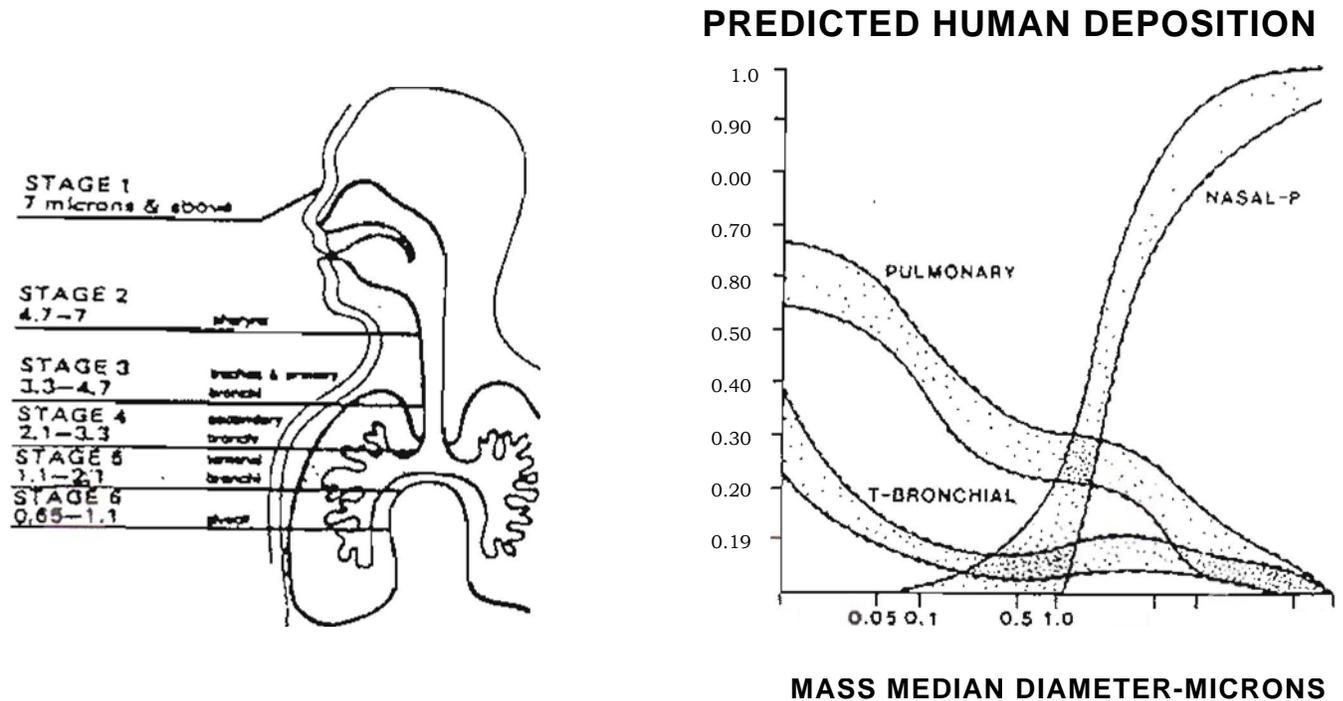


Figure 1 • BGI Sampler simulates Human Respiratory System

After over 50 years of use and application the functioning of a cascade is now accepted as common engineering knowledge. The performance of an impactor is described by the impaction parameter K.

$$\frac{C_p U D_p^2}{18 \mu D_c}$$

Where U is the relative velocity, p is the particle density, D_p is the particle diameter, μ is the gas viscosity, D_c is the diameter of the round jet, and C is the Cunningham slip correction factor.

Data from inertial impactors are normally presented as the 50% effective cutoff diameters. For BGI Impactors, containing round jets and flat collection surfaces, the 50% effective cutoff diameter would yield the value of 0.14 for the inertial impaction parameter K.

The Cunningham slip correction factor (C) is equal to: $1 + 0.16 \times 10^{-4} / D_p$ for normal temperatures and pressures. This factor corrects for the fact that as particle diameters approach the mean free path length of the gas molecules, they tend to "slip" between gas molecules more easily and are more easily able to cross the bulk flow streamlines. Therefore, collection efficiency is slightly greater than would be predicted by inertial impaction theory for particle diameters on the order of 1 or 2 micrometers. The overlapping of particle size between stages, (which is naturally inherent in all cascade impaction devices), can be minimized by design. As a particle passes through a jet, its nearness to the axis of the jet is one of the factors that determines whether or not the particle will reach the impacting surface. The overall performance of multi-orifice impactors is the main reason why they have remained in the forefront of impactor design and use for over 50 years.

Six- Stage Viable Particle Sampler Description

The BGI 1CFM Viable Six Stage Particle impactor is constructed with six aluminum stages that are held together by three spring clamps and sealed with O-ring gaskets (Figure 2). Each impactor stage contains multiple precision drilled orifices. When air is drawn through the sampler, multiple jets of air in each stage direct any airborne particles toward the surface of the agar collection surface for the stage. The sizes of the jet orifices are constant within each stage, but are smaller in each succeeding stage. The range of particle sizes collected on each stage depends on the jet velocity of the stage and the cutoff of the previous stage. Any particle not collected on the first stage follows the air stream around the edge of the Petri dish to the next stage.

Each stage contains 400 orifices with diameters ranging from 1.18 mm on the first stage to 0.25 mm on the sixth stage. Each stage has a removable glass Petri dish with a metal cover. The exhaust section of each stage is approximately 19mm larger in diameter than the Petri dish, which allows un-impacted particles to go around the dish into the next stage.

The Six-Stage Viable Particle Sampler and vacuum pump include their own carrying case for ease and portability.

A constant air sampler flow of 1CFM is provided by a continuous duty vacuum pump. An adjustable valve on the pump controls flow rate and periodic flow rate calibration is recommended.



Figure 2 BGI Six-Stage Viable Sampler
The jet orifice dimensions and particle size ranges for each stage are:

Stage	Orifice Diameter (mm)	D ₅₀	Range of Particle Sizes (Micrometers)
1	1.18	5.8	7 and above
2	0.91	4.7	4.7 – 7.0
3	0.71	3.3	3.3 – 4.7
4	0.53	2.1	2.1 – 3.3
5	0.34	1.1	1.1 – 2.1
6	0.25	0.7	0.65 – 1.1

Assembly

The orifice stages should be cleaned and disinfected each time the instrument is used. A mild detergent and warm water are sufficient for cleaning. The soap can be removed by holding the stages under hot running water or immersing them in clean water in an ultrasonic cleaner. Each stage should be examined for any material in the jet holes. If holes are plugged, or partially plugged, a jet blast of clean dry air is effective in cleaning them. Just before use, wipe all surfaces with 70% isopropyl alcohol using a gauze pad.

The complete impactor assembly consists of an inlet cone and six stages. The stages are inscribed 1, 2, 3, 4, 5, and 6. Each stage contains a silicone O-ring for sealing. These O-rings should be checked regularly and replaced when they no longer provide an airtight seal, part number BGI10706. The assembly of the six-stage impactor begins by placing an agar collection plate (Petri dish), uncovered, on the base plate so that the Petri dish rests on three raised metal pins. Insert stage 6 over the Petri dish. Place a second Petri dish on top of stage 6 and continue this procedure until all six agar collection plates have been positioned in the sampler. The inlet cone is placed on top of stage 1. All the agar plates should be at room temperature before they are inserted into the Sampling Instrument. Hold the inlet cone and plates steady and pull upward on each of the three springs to complete the assembly.

When the glass Petri dishes supplied with the sampler are prepared with 27ml of agar, the three metal pins on each stage position the collection surface for the correct distance between the jet orifices and the agar surface.

After the sampler has been assembled, connect the outlet hose barb on the base plate to the vacuum pump or other vacuum source.

Sampling

When ready to sample, the vacuum pump is turned on and a sample flow rate of 1 ACFM (28.3 LPM) will flow through the sampler. Figure 3 shows how impaction occurs at the orifice collection interfaces.

Normal sampling periods for the viable aerosols will vary from a few minutes up to 30 minutes depending on the purpose for which the sample is being collected and the type of air environment being sampled. It is important to collect sufficient viable particles in each sample to be statistically significant and representative. Difficulty will be experienced in trying to count agar plates, which contain more than 250-300 colonies.

Flow of high velocity sample air across the agar plates also tends to dehydrate and damage the viable particles, which have been collected. Extended sampling periods (over 30 minutes) are not recommended. If a larger sample volume is required, it is better to use two sampling impactors sequentially or to remove the agar plates representing one- sample and insert fresh plates for a second sample.

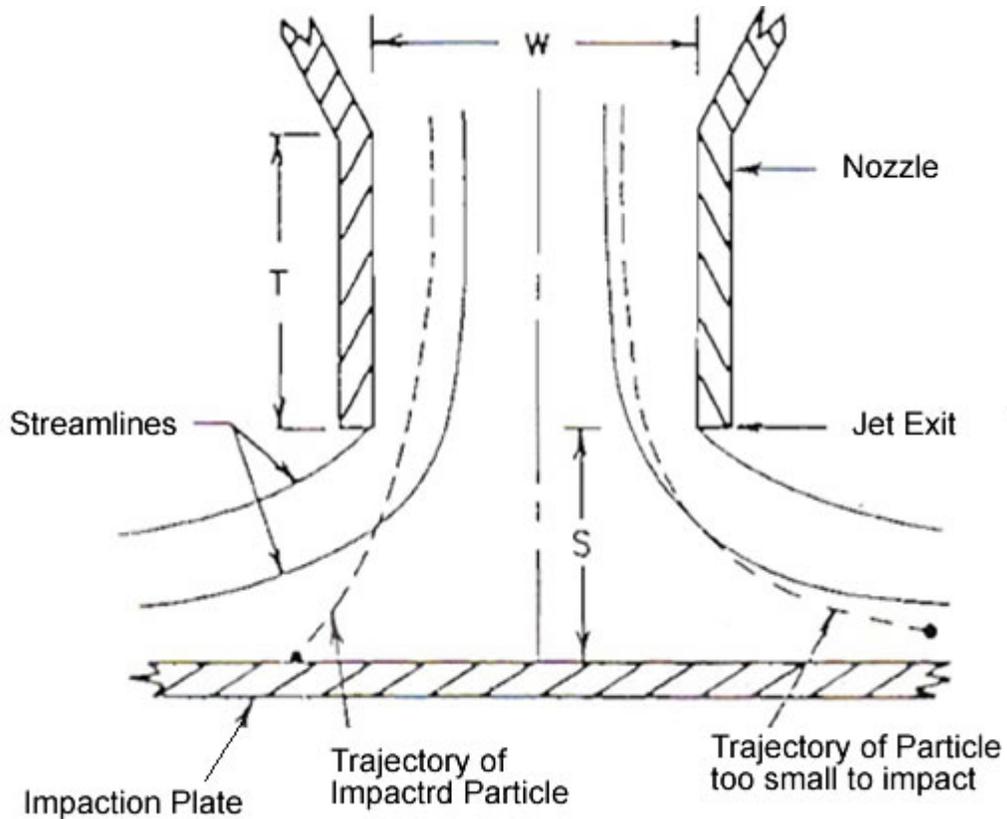


Figure 3 • Schematic of Impactor Stage

After the sampling has been completed, the Sampler is disassembled and the covers are replaced on each of the Petri dishes.

Flow Rate Calibration

Since the orifice velocities determine the size fraction for each stage, it is important that the sampler be operated at 1 ACFM. (28.3 LPM) For this reason, the unit should be periodically recalibrated and whenever non-standard temperatures and pressures are encountered, calibration should be performed at the sampling conditions. Do not use rubber tubing of smaller diameter or length different than that supplied with the impactor, unless the flow rate is readjusted.

Each BGI pump is equipped with an adjustable valve. Always tighten the lock nut on the adjustment valve after the flow rate has been set. To adjust the flow, turn the screw in, to increase flow and out to decrease flow.

Each BGI pump-impactor assembly is calibrated before shipment to deliver 1 ACFM at ambient temperature and pressure levels in Boston, MA, In order to recalibrate at your sampling environment, the following procedure is recommended.

- Place a calibrated Rotameter or electronic flow meter upstream of the sampler. Attach a short 1" I.D. hose with approximately 1/4" wall, to the inlet cone of the impactor and the other end to the calibration device.. Adjust the pump valve until the meter indicates 1 ACFM (28.3 ALPM) then tighten the lock nut on the adjustment valve.

Because of the 1.4 ACFM “free flow” rate of the motor and pump, up to 50 feet of vacuum tubing can be used between the Impactor and pump while maintaining 1 ACFM through the sampler. Note, the impactor must be flow rate calibrated to its specific tubing length.

The pump and motor are guaranteed by the original manufacturer and should not be disassembled for any reason. The vacuum pump uses dry rotary vanes without lubrication. The exhaust jar should be periodically cleaned out from the wear of the fiber vanes. For DC applications contact the BGI factory.

BGI10890
Single Stage Viable (Microbial) Impactor, Terminology NIOSH6 (N6)

Not all Aerobiology sampling requires a complete distribution analysis. There are many simple safety checks or survey activities which can be effectively addressed with less information. For these applications the one (single) stage viable impactor will be used. The characteristics of this single stage impactor is provided in the following table. The single stage impactor comprises only stage number six of the six stage cascade impactor and the base plate with hose barb. A single Petri dish with agar is placed on the base plate and covered with the single number six impactor plate and inlet cone. In all other respects the instructions for the full cascade impactor may be used for the operation of this unit.

Stage	Orifice Diameter (mm)	D ₅₀	Range of Particle Sizes (Micrometers)
1 (6)	0.26	0.7	0.65 - 1

Revision History

V. 1.0.0 Text Created

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