Biological Efficiency Testing of the bioMérieux *air IDEAL®3P™* air sampler following the ISO 14698-1 standard versus the main commercially-available air samplers

bioMérieux Industry Culture Media Group, Chemin de l'Orme, 69280 Marcy L'Etoile, France

air IDEAL®3P™ was third party validated by the Health Protection Agency (UK) to meet the requirements of ISO 14698-1 for the control of clean rooms. This document summarizes and discusses the report N° 988-05, (dated 9th December 2005) from the Health Protection Agency. The full version of the report is available during inspection.

Abstract:
The bioMérieux air IDEAL®3P™ microbial air sampler has been tested for biological efficiency following the ISO 14698-1 standard (ref. 1). The sampler was shown to be able to collect aerosols of *Staphylococcus epidermidis* with a high efficiency (92.1%) compared to the gold standard Casella slit sampler. The collection efficiency of *S. epidermidis* and *B. subtilis*. var. *niger* was also found to be higher than those of three main commercially-available air samplers.

Material and methods:
Test sampler: the air IDEAL®3P™ air sampler (sampler A) is an impactor type of instrument based on the principle described by Andersen *et al.* (ref. 2), in which air is aspirated through a grid perforated with a pattern of 265 calibrated holes. The resulting air streams containing microbial particles are directed onto the agar surface in a bioMérieux irradiated Trypcase Soy Agar plate.

Reference sampler: Casella slit sampler calibrated operating at 30L/min. This non-portable sampler is known to collect bacteria with high efficiency and is used as the gold standard for air sampling evaluations.

Commercially-available air samplers: three air samplers based on the principle of impaction using irradiated Trypcase Soy Agar (TSA) recommended by the different instrument manufacturers.
•Instrument B: Merck MAS 100. Sampler operating at 100L/min and using TSA 90mm irradiated plates (ref AXO51146 from Merck).
•Instrument C: Biotest RCS Plus. Sampler operating at 50L/min and using TSA irradiated strip (ref 941115 from Biotest).
•Instrument D: PBI SAS Super 100. Sampler operating at 100L/min and using TSA 55 mm irradiated plates (ref 103070 from Redipor). In this case the instrument manufacturer has no special recommendation concerning the media.

Spray suspension: mixed microbial suspension of *B. subtilis* var *niger* and *S. epidermidis.* The *B. subtilis* spores are recognized as an aerostable microbial tracer and thus, used as indicator of the physical efficiency of the air samplers. *S. epidermidis* is a common contaminant of the air in pharmaceutical clean-rooms derived from human skin cells. This strain was used as the test micro-organism for the biological efficiency testing.

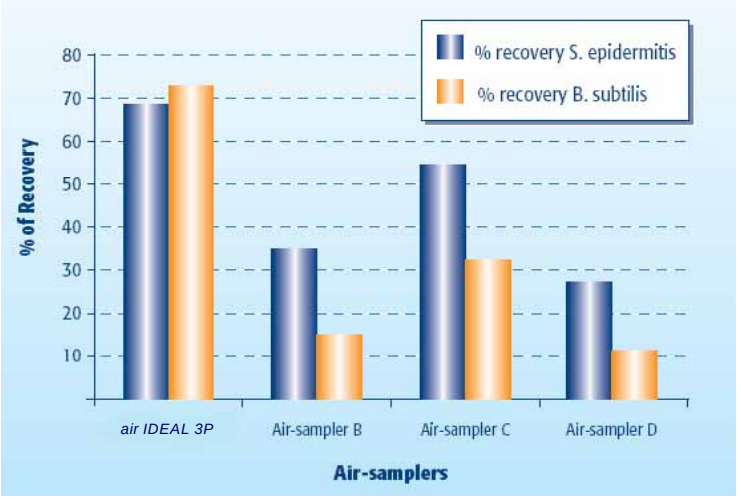
Aerosol generation: A three jet Collison nebuliser (ref. 3) operated at a pressure of 26 psi was used to generate the mixed microbial aerosol. This suspension was sprayed in a 18m^3 environmental chamber.

The air content of the room was sampled simultaneously by the air IDEAL®3P™, the three commercially-available air samplers and the reference sampler. Each measure was taken 22 times. After incubation at 37°C the small white colonies of *S. epidermidis* (SE) and the larger orange colonies of *B. subtilis* (BS) were counted individually. The biological efficiency of air IDEAL®3P™ was calculated as follows:

$$\text{Biological efficiency} = \frac{\text{air IDEAL®3P™'s ratio SE/BS} \times 100}{\text{reference sampler's ratio SE/BS}}$$

In order to evaluate the physical and biological efficiencies of air IDEAL®3P™ versus the main commercially available air samplers, a percentage of recovery of both strains, for each of the 22 runs, was calculated using the Casella sampler as reference. The figure 1 shows the average percentage of recovery of each air sampler obtained with *B. subtilis* and *S. epidermidis*.

Fig. 1 Collection efficiency of air IDEAL 3P vs main commercialy available air-samplers



Results and Discussion:
The effectiveness of microbial air samplers can be split into two facets: physical and biological efficiencies. Physical efficiency is the ability of the sampler to collect airborne particles of various sizes, while biological efficiency is the ability of the sampler to collect airborne micro-organisms without rendering them non-viable. The physical efficiency of the air IDEAL®3P™ has already been tested by the HPA (report number 970-05) and has demonstrated that air IDEAL®3P™ has a high level of collection of the particles of interest.

This study highlights that **there is no significant difference between the biological efficiency of air IDEAL®3P™ and the gold standard Casella sampler** in the 22 tests (p=0.234, paired Student t-test) with an overall comparative biological efficiency of 92.1% (data not shown).

Furthermore, this study demonstrates the **superior performances of this new instrument compared to the main commercially-available air samplers** on both reference strains tested: *B. subtilis* (indicator of physical efficiency) and *S. epidermidis* (indicator of biological efficiency). The low recovery observed in these experiments with competitive air samplers could potentially be linked to a low physical efficiency of these instruments. Another explanation that could play a role is the quality of the media and their compatibility with each instrument.

1. NF EN ISO 14698-1 "Clean rooms and associated controlled environments – Biocontamination control". (2004)
2. Andersen, A.A. "New sampler for the collection, sizing and enumeration of viable airborne particles." J. Bacteriology. (1976).
3. May, KR. "The Collison nebuliser: Description, performances and application". Aerosol Science. (1973).