# Comparative study of Exhaust Air Dust (EAD®) testing and traditional bedding sentinels for health monitoring in IVC rack systems

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## 1 INTRODUCTION

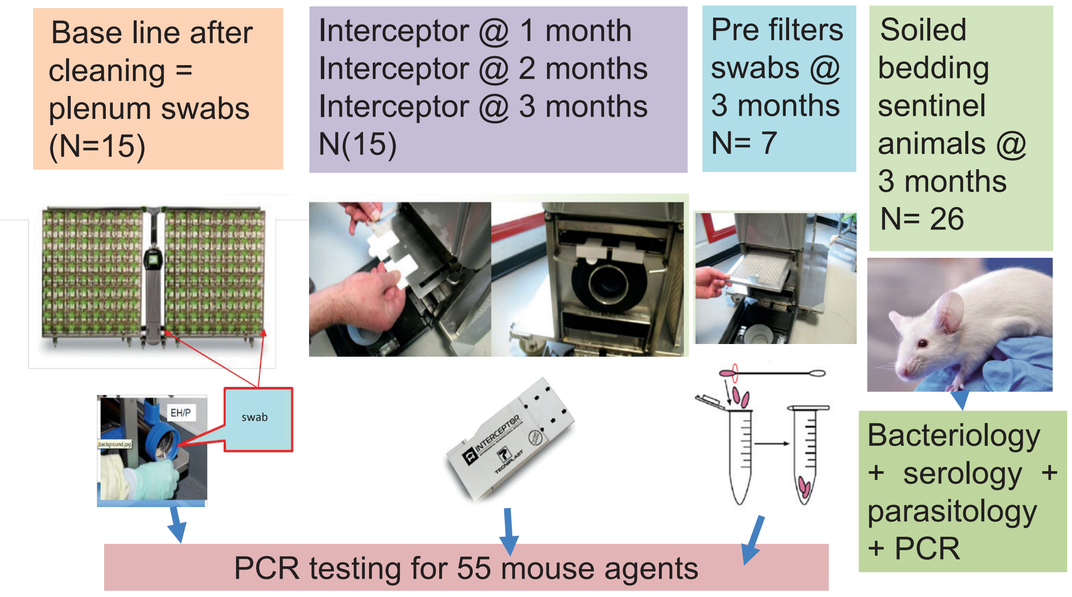
Alternative health monitoring such as Exhaust Air Dust (EAD®) PCR testing is a new method designed to improve the detection of pathogens. EAD® can be easily collected with in-line capture media available in different IVC rack systems.

Three independent research groups (Max Planck Institute-Frankfurt; Helmholtz Zentrum- Munich, Cogentech Research Center- Milano) performed a health monitoring comparison study.

They compared the accuracy of environmental health monitoring by using an in line EAD® filter in IVC racks (N=26) to the traditional bedding sentinel methodology.

## 2 MATERIAL AND METHODS

**Figure 1:** Sample collection




Each facility cleaned between 2-5 air handling units (AHU, Tecniplast) with different types of rack clusters (between 1 to 4 single or double racks linked to the AHU). In total, the study included 9 AHU's with 26 IVC racks. Each sample type was pre-processed and DNA/RNA was extracted using a total nucleic acid isolation kit. Sample nucleic acid was screened by individual single-plex PCR assays on a real time open array PCR platform. Semi quantification was performed to define copy number per reaction. Test specimens at 1, 2 and 3 months included base line swabs, prefilter swabs (N=7) and in-line EAD® filters (Interceptor, N=15) for environmental samples. At 3 months, soiled bedding sentinels (N=26) were sent to laboratories for traditional testing including bacteriology, serology, parasitology and PCR (Fig. 1).

## 3 RESULTS

Post cleaning, baseline PCR tests were all negative except for one sample which was found positive for *Pasteurella pneumotropica* at very low copy number (1-10 copies)(data not shown).

**Table 1**: Percentage of detected agents

| Agents | Soiled bedding sentinels | Interceptor (N=15) | Prefilter swabs (N=7) |
|---|---|---|---|
| Virus | | | |
| MNV | 42,3% | 46,7% | 42,86% |
| bacteria | | | |
| Pasteurella pneumotropica-Jawetz | 3,8% | 53,3% | 85,71% |
| Pasteruella pneumotropica-Heyl | 0,0% | 60,0% | 57,1% |
| Helicobacter genus | 23,1% | 100,0% | 100,0% |
| Helicobacter hepaticus | 7,7% | 60,0% | 85,7% |
| Helicobacter ganmani | 0,0% | 80,0% | 100,0% |
| Helicobater mastomyrinus | 0,0% | 46,7% | 57,1% |
| Helicobater typhlonius | 0,0% | 73,3% | 100,0% |
| Pseudomonas aeruginosa | 7,7% | 0,0% | 42,9% |
| Staphylococcus aureus | 0,0% | 20,0% | 28,6% |
| parasites | | | |
| Myocoptes musculinus | 0,0% | 6,7% | 14,29% |
| Entamoeba | 3,8% | 40,0% | 28,57% |
| | | | |
| Average | 7,4% | 48,9% | 61,9% |

Bad — Good

The data showed a significant increase in detection rate with the filter media. Overall, the average percentage of detected agents using the media filter was 7 time higher as compared to the sentinels (Table 1).

Most of the agents were detected after 1 month of exposure (Fig. 2). Agents such as *S. aureus, P. aeruginosa* showed a delayed detection at 2 or 3 months (Fig.2).

*Helicobater spp* showed the highest copy number where as *S. aureus* showed the lowest copy number (Fig.3). Opportunistic agents such as *S. aureus* shedded at a very low rate in immunocomptent mice.

**Figure 2**: Percentage of detected agents

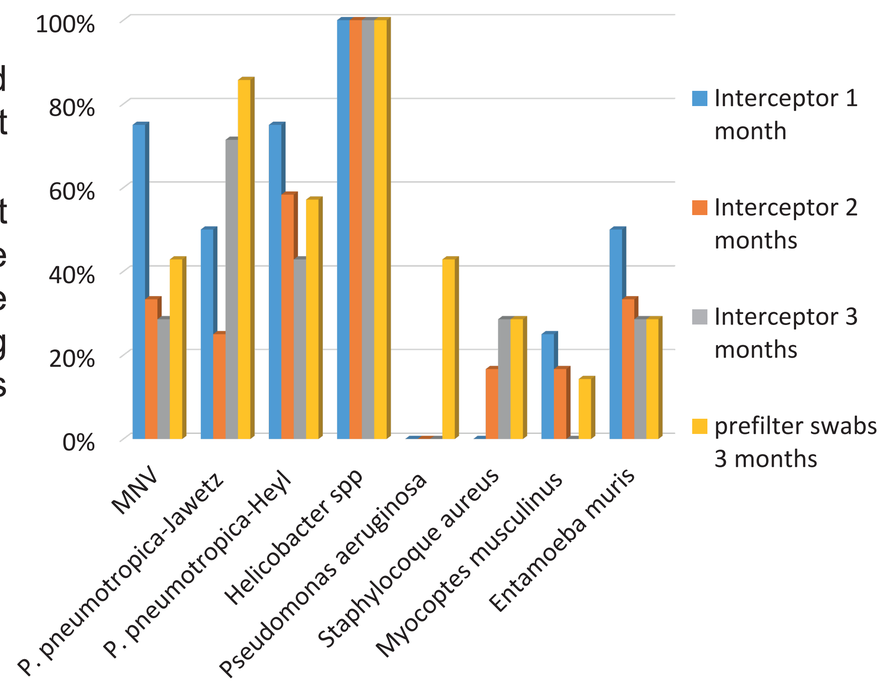
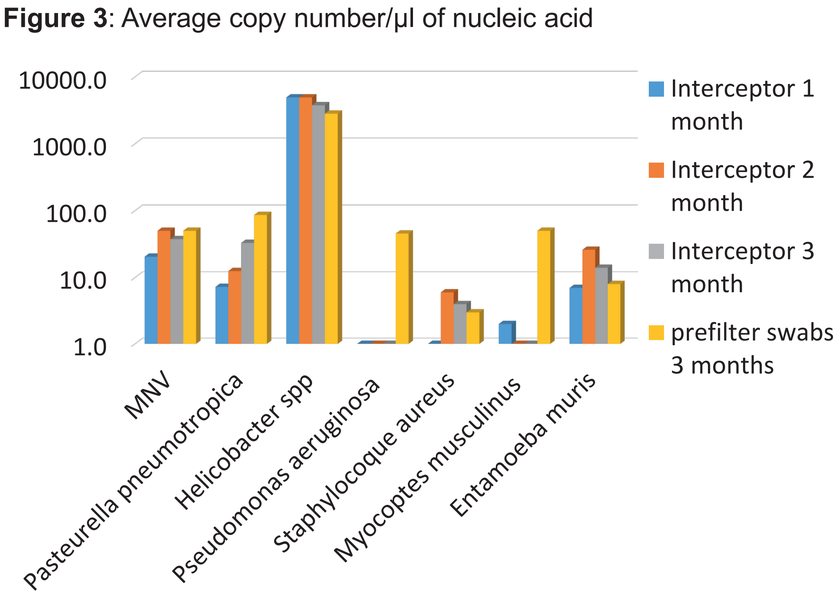



**Figure 3**: Average copy number/µl of nucleic acid




## 4 CONCLUSION

- EAD® testing is an effective method for health monitoring: it improves the detection of pathogens as compared to soiled bedding sentinels in IVC racks
- EAD® testing can help reduce the number of sentinel animals needed per experiment: It's a positive way to apply the 3R's in an animal facility.

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