Prof. Dr. med. W. Solbach
Direktor des Instituts für
Med. Mikrobiologie und Hygiene
Universität zu Lübeck

Campus Lübeck

Ratzeburger Allee 160
23538 Lübeck

Prof. Dr. med. W. Solbach • Ratzeburger Allee 160 • 23538 Lübeck

Diagnostische Laboratorien
Auskunft: 0451 500-2824
akkreditiert nach DIN EN ISO 15189:2007 und
DIN EN ISO/IEC 17025:2005

Deutscher Akkreditierungs Rat
DAR
DGA-ML-6157.02.10

14.02.2014

Microbiological assessment of the breath alcohol measuring device Dräger Interlock® 5000/7000

1. Background

Several countries require cars to be equipped with breath alcohol ignition interlock devices (BAIID), e.g. vehicles used for transportation of school children in Finland. If the BAIID detects breath alcohol above a set point, the vehicle's engine cannot be started. BAIIDs may also be installed in private vehicles as a part of alcohol offender programs (Sweden, USA).

Devices used by more than one person may pose a risk of transfer of infectious agents between the users. The assessment of such a risk is therefore deemed necessary. In 2006, the Institute of Med. Microbiology and Hygiene assessed the current device's predecessor, the "Dräger Interlock XT". The XT mouthpiece's non-return valve has been discontinued for the new device's mouthpiece, requiring a new assessment of the device's microbiological status.

2. Methods

2.1 Simulation "Usage of BAIID"

Between 06.09.2013 and 16.09.2013, the BAIID was tested for 10 days. Instead of being installed in a car, the device was connected to mains using an external power supply, thus the device was not continuously heated. During the test period, a test subject provided two breath samples per day, one in the morning and one in the afternoon. Following each breath sample, the device was left turned on, and thus heated, for another hour. On the last day of testing, a breath sample was collected in the morning, and the device was left heated for four hours. After four hours it was immediately used for the following microbiological tests.

2.2 Microbiological experiments

All parts were removed under a sterile workbench.

All microbiological analyses of parts of the BAIID that were used during the simulation are considered as positive samples. Parts from a new, unused but openly stored BAIID were used as negative controls. Plating was performed on solid culture media blood agar and chocolate agar. All streaks were evaluated in repeat determination.

Agar dishes were incubated aerobic at 36 °C for 72 hours.

Chocolate agar dishes were incubated at 36 °C for 72 hours in 5% CO_2.atmosphere.

The following parts of the BAIID were evaluated (see Figure 1)

- Mouthpiece (not shown)
- Tube, leading to the pressure sensor
- Transition from mouthpiece to the tube (AS)
- Air outlet on the BAIID's front side (AL)

2.2.1 Mouthpiece
The mouthpiece was added to a 500 ml round-flask together with 200 ml CASO boullion, and shaken at 150 rpm for 120 min at room temperature. 0.1 ml each were streaked onto agar plates straight from the preparation (0) and from two dilution steps (10^{-1}, 10^{-2}).

2.2.2 Tube, unheated
The device's housing was opened, the tube was removed with a sterile pair of tweezers. After disinfecting and drying the tube's outer side, it was added to a test tube filled with 10 ml CASO boullion, vortexed, settled for 120 min at room temperature and prior to pipetting on to agar-plates vortexed again. 0.1 ml each were streaked onto agar plates straight from the preparation (0) and from two dilution steps (10^{-1}, 10^{-2}).

2.2.3 Transition from mouthpiece to the tube.
A sterile swab, wetted with NaCl solution, was used to wipe the area between the mouthpiece and the opening to the tube several times. The swab was added to a testing tube filled with 10 ml CASO boullion, vortexed, settled for 120 min at room temperature and prior to pipetting on to agar-plates vortexed again. 0.1 ml and 0.5 ml were streaked out straight from the preparation.

2.2.4 Air outlet on the BAIID's front side
A sterile swab, wetted with NaCl solution, was used to wipe the area between the mouthpiece and the air outlet several times. The swab was added to a testing tube filled with 10 ml CASO bullion, vortexed, settled for 120 min at room temperature and prior to pipetting on to agar-plates vortexed again. 0.1 ml and 0.5 ml were streaked out straight from the preparation.

3. Results
3.1 Mouthpiece
On the negative mouthpiece, there were no or only trace amounts of germs detectable, as expected (see Table 1).

The used positive mouthpiece was highly contaminated. Extrapolated to the complete mouthpiece, using an aerobic incubation a mean germ number (colony forming units; CFU) of 20,100 was found; under 5% CO_2 incubation, the mean germ count was 17,600 (extrapolation from the initial preparation (0).

	CFU on blood agar		
Dilution	0	10^{-1}	10^{-2}
Mouth piece positive	93 / 108	7 / 6	7 / 6
Mouth piece	0 / 3	0 / 0	0 / 0

negative			
	CFU on chocolate agar, 5% CO_2		
Mouth piece positive	75 / 101	3 / 10	3 / 1
Mouth piece negative	0 / 0	0 / 0	0 / 0

Table 1

Identification:
Colonies found on the positive mouthpiece were identified using MALDI-TOF spectrometry. Detectable were germs ususally found in the human flora of nose, mouth and faeces like *Staphylococcus aureus, Heamophilus parainfluenzae, Eikonella corodens, Neisseria macacae, Streptococcus oralis,* and *Streptococcus pneumoniae*. Additionally, spore forming germs (airborne) were found. The negative mouthpiece showed micrococcus.

3.2 Tube, unheated

	CFU on blood agar		
Dilution	0	10^{-1}	10^{-2}
tube positive	0 / 0	0 / 0	0 / 0
tube negative	0 / 0	0 / 0	0 / 0
	CFU on chocolate agar, 5% CO_2		
tube positive	0 / 0	0 / 0	0 / 0
tube negative	0 / 0	0 / 0	0 / 0

Table 2

3.3 Transition from mouthpiece to the tube
On the area leading from the mouthpiece to the tube, both devices (used/unused) showed germs in small numbers (less than 100 colony-forming units per streak).

	CFU on blood agar	
	CFU/0,1 ml	CFU/0,5 ml
output transition mouthpiece - tube positive	0 / 0	0 / 1
output transition mouthpiece - tube negative	1 / 0	1 / 2
	CFU on chocolate agar, 5% CO_2	
output transition mouthpiece – tube positive	0 / 0	0 / 0
output transition mouthpiece – tube negative	0 / 0	4 / 3

Table 3

Identification:
Only micrococcus were detectable. These are germs are naturally occurring in the flora of the skin.

3.4 Air outlet on the BAIID's front side

In contrast to the negative BAIID, germs were detected on the positive BAIID's air outlet; around 1,500 colony-forming units could be found there.

	CFU on blood agar	
	CFU/0,1 ml	CFU/0,5 ml
output air outlet positive	16 / 14	80 / 76
output air outlet negative	0 / 0	0 / 0
	CFU on chocolate agar, 5% CO_2	
output air outlet positive	14 / 16	66 / 64
output air outlet negative	0 / 0	0 / 0

Table 4

Identification:
On the air outlet, coagulase-negative staphylococcus was detected. These are germs are naturally occurring as part of the skin microbiota.

Conclusion:
The largest contamination could be found on the used mouthpiece. The germs found were germs found in the human flora of nose, mouth and fauces. Air leading systems showed much less contamination, and only contamination from germs found on the skin.

There is no concern against using the BAIID under test in the field. Every user should use its own mouthpiece. A wipe-disinfection is recommended when the user changes.

Professor Dr. med. Werner Solbach