BIOAEROSOL EXPOSURE IN WASTE COLLECTION: A COMPARATIVE STUDY ON THE SIGNIFICANCE OF COLLECTION EQUIPMENT, TYPE OF WASTE AND SEASONAL VARIATION

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Abstract—Recent Danish studies on waste collectors' bioaerosol exposure are summarized. Generally the median exposure levels ranged from $10^5$ to $10^6$ cells m$^{-1}$ (total microorganisms), $10^4$ to $10^5$ cfu m$^{-1}$ (culturable fungi) and $10^2$ to $10^3$ cfu m$^{-1}$ (culturable bacteria). The type of waste was a governing factor for exposure. Garden waste collectors frequently experienced concentrations exceeding $10^5$ cfu m$^{-1}$ for mesophilic fungi and $10^4$ cfu m$^{-1}$ for the thermophilic fungus *Aspergillus fumigatus*. Workers collecting compostable, mixed and sorted waste occasionally experienced similar concentrations of the fungal groups while workers collecting 'bulky waste' and paper had low exposure. Type of collection vehicle was identified as another governing factor for exposure. Vehicles loaded from the top (approximately 3 m above the ground) caused lower exposure (by a factor of 25) to fungi than vehicles loaded at the level or the breathing zone of the workers. Exposure was also affected by season of the year—the concentration of total microorganisms, culturable fungi, *Aspergillus fumigatus* and endotoxin was low in winter. Exposure to total microorganisms counted by microscopy was found to have a fairly high validity ($V_a$) as an indicator of exposure to culturable fungi ($V_a = 1.45$) or culturable bacteria ($V_a = 1.25$). Likewise, dust may also be used as an indicator of exposure to total microorganisms ($V_a = 1.36$), culturable fungi ($V_a = 1.31$) and culturable bacteria ($V_a = 1.35$). © 1997 British Occupational Hygiene Society. Published by Elsevier Science Ltd

INTRODUCTION

In recent years much effort has been put into the recycling of household waste. To improve the quality of the recovered materials recycling at the household level often involves pre-separation of the waste into different fractions including compostable kitchen waste, garden waste, paper and glass. The segregation of household waste creates differences in the microbiological potential of the different waste fractions, and for collecting segregated waste new designs of containers and trucks are introduced. These new systems are likely to influence the occupational environment for the workers collecting the waste. As handling of waste may cause microorganisms and dust to become aerosolized, waste collectors are at risk of being exposed to bioaerosols generated from the waste. For waste collectors the bioaerosol exposure probably depends on such factors as the microflora of the waste, the type of container, the truck and the organization of work. Segregation of
household waste may therefore cause some waste collectors to be more heavily exposed to bioaerosols than others. However, data on waste collectors’ bioaerosol exposure are sparse (Poulsen et al., 1995a,b). Therefore the Danish Environmental Protection Agency and the Danish Work Environment Service jointly initiated a 5-year (1994–1998) research programme (‘Waste Collection and Recycling’) with the focus on occupational exposures and adverse health effects in relation to collection, sorting and recycling of the household waste. So far the programme has concentrated on waste collectors and the data obtained on their bioaerosol exposure are summarized in the present paper.

Waste collectors’ bioaerosol exposure may be correlated with several factors including the type of waste, season of the year, equipment at the households, type of vehicle used for collection, and organisation of work. However, knowledge on exposure against governing factors is limited and the purpose of the present paper is to characterize waste collectors bioaerosol exposure in relation to these factors. The paper also introduces the concept of total dust or total counts of microorganisms (live and dead) as indicators of exposure to viable fungi or bacteria.

MATERIALS AND METHODS

Types of waste and equipment for collection

Data from eight individual studies of personal bioaerosol exposure during waste collection are summarized. The types of containers, trucks and waste are listed in Table 1. Mixed household waste is defined as unseparated waste generated at private homes. Compostable household waste or the biodegradable fraction is mostly food remains. The household waste of study IV was sorted into two fractions at the households: the compostable fraction and the ‘rest’ fraction. These fractions were stored separately in two-compartment containers. Garden waste consists of all sorts of compostable waste from private gardens, including branches, leaves, grass, rotten fruit, and so on. The ‘bulky waste for incineration’ in study VIII includes furniture and large pieces of wood. Study VIII also included the collection of different fractions of recyclable material: paper in bundles, glass, metal, cardboard, and plastic.

A number of different truck designs for waste collection are in use in Denmark. The most widely used compactor truck has a closed container for storage of the waste, and at the rear a lift for automatic emptying of bins and waste containers into a magazine (the scoop) fitted to the container of the truck (low opening, approximately 1.4 m above the ground). When loading sacks into compactor trucks, the worker lifts the waste sack manually into the scoop. To enhance the speed of emptying, the bins are also often emptied manually instead of using the lift. When the scoop is full, the waste is mechanically pushed into the container and compacted. A ‘bio-truck’ has a closed container without a compression mechanism, but with a lift for loading from the top of the container (approximately 3 m above the ground). When a platform truck is used for collection of waste in sacks, the sacks are thrown up and stacked manually on the open truck body. For collection of various fractions of recyclable materials, the truck body is divided into sections.

In most of the studies, the waste collectors worked in crews of two to three men operating one truck. If not otherwise stated, all crew members participated in all
Table 1. Description of the waste, truck, equipment at the household and the season where bioaerosol sampling took place

<table>
<thead>
<tr>
<th>Study</th>
<th>Waste</th>
<th>Truck</th>
<th>Equipment</th>
<th>Season</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>mixed household (7 days)*</td>
<td>compactor</td>
<td>4-wheel containers (400, 600 l.)</td>
<td>August</td>
</tr>
<tr>
<td>II</td>
<td>mixed household (7 days)</td>
<td>compactor (a) basic (b) exhaust compactor</td>
<td>2- and 4-wheel containers (110, 300, 400 and 600 l.)</td>
<td>June–July</td>
</tr>
<tr>
<td>III</td>
<td>mixed household (a) 7 days (b) 14 days</td>
<td>compactor (a) (b) compactor</td>
<td>(a) bins (150 l.) (b) 2-wheel containers (240 l.)</td>
<td>August–September</td>
</tr>
<tr>
<td>IV</td>
<td>(a) sorted household waste (7 days) (b) paper (14–30 days)</td>
<td>compactor (a) high loading (b) basic compactor</td>
<td>(a) 2-compartment containers (230 l.) (b) 2-wheel containers (190 l.)</td>
<td>August</td>
</tr>
<tr>
<td>V</td>
<td>compostable household (14 days)</td>
<td>compactor (a) compactor (b) compactor (c) platform truck</td>
<td>(a) 2-wheel containers (110 l.) (b) paper sacks (90 l.) (c) paper sacks (90 l.)</td>
<td>spring, summer, autumn, winter</td>
</tr>
<tr>
<td>VI</td>
<td>compostable household (14 days)</td>
<td>compactor (a) compactor (b) special design compactor</td>
<td>bio-truck</td>
<td>June</td>
</tr>
<tr>
<td>VII</td>
<td>garden waste</td>
<td>2-wheel containers and unpacked in bundles</td>
<td>special design compactor</td>
<td>November</td>
</tr>
<tr>
<td>VIII</td>
<td>(a) garden waste (b) bulky waste for incineration (c) paper, glass, etc. (1 month)</td>
<td>compactor (a) compactor (b) compactor (c) platform truck</td>
<td>(a) plastic sacks and unpacked in bundles (b) no packing (c) bundles or bags, boxes, etc.</td>
<td>May, October</td>
</tr>
</tbody>
</table>

*Frequency of collection (age of waste).

waste handling tasks. Except for study V, the workers had a fixed wage per day but were allowed to go home when the waste was collected in the catchment area. The specific details of the different studies are described below.

Study I. The investigation (Nielsen et al., 1995a) took place in a city area of multistorey houses. Samples were collected in two consecutive weeks (8 sampling days). Full-shift bioaerosol samples were obtained from a single crew of three waste collectors. Each member of the crew had a specific task: the ‘runner’ operated ahead of the truck by moving containers from the house to curbside; the ‘loader’ emptied the containers into the truck and brought them back; and the ‘driver’ drove the truck but sometimes also assisted the loader. These tasks were performed alternately by the three workers during the week.

Study II. Sampling took place in two city districts (multistorey houses) selected to be as similar as possible. Bioaerosol samples from two crews, operating two different types of compactor trucks, were simultaneously collected once a week in the districts (Breum et al., 1996a). One truck was a standard compactor truck as described above. The other was a standard compactor truck fitted with a sliced plastic curtain covering part of the air space above the scoop and with an air exhaust system. From behind the curtain the air was exhausted from an outlet at the centre of the roof above the scoop.
Study III. Sampling took place in two districts with mainly one-family houses. In one district the waste, mainly in two-wheel containers, was collected every fortnight. In the other, the waste kept in bins was collected once a week. Bioaerosol samples were collected simultaneously from the two crews operating in these districts (Breum et al., 1995b).

Study IV. Sampling took place in districts with multistory houses or one-family houses (Würtz et al., 1995). The households sorted the waste into two fractions: the compostable fraction and the 'rest' fraction. The two fractions were stored separately in an out-door two-compartment container. Special compactor trucks with a two-compartment body was used for collecting the waste. The truck was loaded from approximately 3 m above the ground. In emptying a household container each waste fraction was automatically directed to the correct compartment of the truck. In addition, the households delivered paper to a separate container.

Study V. The investigation (Nielsen et al., 1995b) took place in several districts of mainly one-family houses in small towns. Bioaerosol samples were obtained during collection of compostable household waste using different types of equipment and during four periods of the year (Table 1).

Study VI. Bioaerosol measurements were made on two systems specially designed for collection of the wet, compostable fraction of household waste. A ‘bio-truck’ was used for emptying bins lined with plastic bags. The truck was loaded approximately 3 m above the ground. In the kitchen, the waste was typically kept in small plastic bags before depositing in the bin. The specially designed ‘bio-truck’ (‘Bates-truck’) had a lift-system for paper sacks and the truck was loaded approximately 3 m above the ground. For this system (the ‘Bates-system’), the waste was kept in small paper bags in the kitchen. It is noted that none of the two types of ‘bio-trucks’ compressed the waste loaded into the trucks. Sampling took place on the same days in two districts of multistorey houses (Poulsen et al., 1995b).

Study VII. This investigation on collection of garden waste took place in districts of one-family houses with gardens in a period of 3 consecutive days (Breum et al., 1995a).

Study VIII. In districts of single-family houses in a large town, the bioaerosol exposure was measured during collection of garden waste, recyclable material (paper, glass) and bulky waste for incineration by the use of different systems (Table 1). A 4-day sampling campaign was used for each system during two different seasons (Breum et al., 1996b).

Bioaerosol sampling

The workers participating in the studies were fitted with personal sampling equipment while they were collecting waste. For a full work period, each worker carried two field monitors for collection of dust (‘total’ particulate matter) and airborne microorganisms. The field monitors were placed in the breathing zone and
connected to portable pumps. According to the Danish standard method (Stubbe Tegbjærg and Wilhardt, 1981) dust was collected on cellulose nitrate filters (25 mm dia., 8 μm; Sartorius, Göttingen, Germany) using closed-face Millipore field monitors (5.6 mm dia. inlet; Millipore, Bedford, U.S.A.) operated at 1.9 l min⁻¹. Samples for analysis of airborne microorganisms were collected on sterile polycarbonate filters in filter cassettes (25 mm dia., 0.4 μm; 4.4 mm dia. inlet; Nuclepore, MA, U.S.A.) operating at an airflow rate of 1.0 l min⁻¹.

Identical sets of sampling equipment were used for area sampling (outdoor reference) at positions near the administration buildings of the waste collecting companies. As a control, blank filters were handled in parallel to the exposed filters in the field and throughout analysis.

**Dust and endotoxin**

The mass of collected dust was determined gravimetrically by weighing the cellulose nitrate filters before and after sampling. The filters were kept at constant air temperature and humidity for a minimum of 24 h before weighing. For analysis of endotoxin the collected dust was resuspended in 10 ml sterile, non-pyrogenic water by orbital shaking (300 rev min⁻¹, 15 min) at room temperature. In studies II–VIII, endotoxin analysis was performed in duplicate by the kinetic Limulus Amoebocyte Lysate test (kinetic-QCL endotoxin kit; BioWhittaker). A standard curve obtained from an *Escherichia coli* 055:B5 reference endotoxin was used to determine the concentrations in terms of endotoxin units (EU) per m³ of air (1 ng = 15.5 EU). In study I, endotoxin was analysed by the quantitative chromogenic Limulus Amoebocyte Lysate test (Coatest Endotoxin; KabiVitrum, Sweden). The standard curve was in this case obtained from an *Escherichia coli* 0111:B4 reference endotoxin (1 ng = 12 EU). The two endotoxin methods should give comparable results in terms of EU.

**Microorganisms**

Microorganisms were quantified by a modification of the CAMNEA-method (Palmgren et al., 1986). Basically, this method involves resuspension of the aerosols collected on the polycarbonate filter, followed by enumeration by epi-fluorescence microscopy (total counts) and by culturing (culturable counts). For the resuspension, 5 ml sterile 0.05% Tween-80 was added to the filter cassette followed by orbital shaking (500 rev min⁻¹) for 15 min at room temperature. Samples for culturable counts were plated immediately when received by the laboratory (up to 20 h after sampling) and those for determination of total counts were kept at −20°C for later examination.

The total number of microbial cells collected on a filter was derived from analysis on a 1.0 ml sample of the extraction fluid or an appropriate dilution of this. The sample was stained with 0.3 ml 0.01% acridine orange in acetate buffer (bioMérieux, France) for 30 s and filtered through a dark polycarbonate filter (25 mm, 0.4 μm; Nuclepore). By epi-fluorescence microscopy (1250× magnification) the number of bacteria and fungal spores were counted in 40 randomly chosen fields or until at least 400 microorganisms had been counted. Live as well as inactivated microbial cells are included by this method. The minimal detectable number was
3x10^3 microorganisms per filter, equivalent to about 10^4 cells m^{-3} of air, depending on the volume of air sampled.

Culturable bacteria and fungi were enumerated by inoculating 0.1 ml of 10-fold dilutions of the resuspension medium on solid media for five groups of microorganisms. For counts of mesophilic bacteria and thermophilic actinomycetes, samples were plated on nutrient agar (Oxoid, Basingstoke, U.K.) supplemented with cycloheximide (Actidione; 50 mg/l), and the plates were incubated for 7 days at 25°C or 55°C, respectively. Ten per cent nutrient agar with cycloheximide (50 mg l.^{-1}) was used for the enumeration of mesophilic actinomycetes (25°C for 7 days). Dichloran Glycerol agar (DG18 agar; Oxoid) was used for the enumeration of mesophilic fungi (25°C for 7 days) and the thermophilic *Aspergillus fumigatus* (45°C for 3 days). For all the media the minimum detectable number of colony forming units (cfu) was 50 per filter, equivalent to approximately 100–200 cfu per m^3 of air. Culturing for mesophilic actinomycetes were not performed in all studies.

**A model for screening exposure to microorganisms**

The more detailed the analysis of microbial content in a sample of bioaerosols required, the more costly and time-consuming is the analytical burden. The use of an indicator for part of the analysis may ease the analytical burden provided that an acceptable sensitivity, specificity and validity of the indicator are achieved. For this presentation first 'total particulate matter' and then total number of microbial cells were used as indicators of exposure to some type of microorganism. Four-fold tables were used for the analysis (Breum and Holst, 1986). One table was used for each indicator and each type of microbial parameter. The basic idea of the analysis was to allow the concentration of a microbial parameter to be classified as 'high' or 'low/intermediate' on the basis of a measured indicator. In this context 'high' was defined as concentrations above the following levels: total microorganisms: 10^6 cells m^{-3}, fungi: 10^5 cfu m^{-3}; bacteria or *A. fumigatus*: 10^4 cfu m^{-3}. These cut-offs were based on data from the literature (see discussion) indicating concentrations of microorganisms that might cause health problems. If the concentration of the indicator exceeded a screening limit (SL) the concentration was classified as 'high'. From the four-fold table (Table 2), the following parameters can be calculated: the frequency of true positives (sensitivity, Se), the frequency of true negatives (specificity, Sp), the validity (Va), the predictive value of a positive screening test.

<table>
<thead>
<tr>
<th>Concentration of the indicator (dust or micro-organisms)</th>
<th>Concentration of some type of micro-organism</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>C &gt; SL</td>
<td>C ≤ L</td>
<td></td>
</tr>
<tr>
<td>C &gt; SL</td>
<td>a</td>
<td></td>
</tr>
<tr>
<td>C ≤ SL</td>
<td>b</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>a + b</td>
<td></td>
</tr>
</tbody>
</table>

L, level for a concentration to be considered 'high' (total microorganisms: 10^6 cells m^{-3}, fungi: 10^5 cfu m^{-3}; bacteria or *A. fumigatus*: 10^4 cfu m^{-3}); SL, screening limit.

Sensitivity, Se = a/(a + c); specificity, Sp = d/(b + d); validity, Va = Se + Sp.

Predictive value of positive result, PV1 = a/(a + b); predictive value of negative result, PV2 = d/(c + d).
result (Pv₁), and the predictive value of a negative test result (Pv₂). By stepwise changing SL, data were obtained on SL against Se, Sp, Va, Pv₁ and Pv₂. The optimum value of SL was chosen by maximizing the validity and require that Se > Sp. The second criterion was used when two optima for Va were found.

Statistical methods

Hypotheses on differences between groups of data were tested non-parametrically by using the Mann–Whitney test. Data were generally presented by median values and range (minimum and maximum values). SAS statistical software (version 6.10; SAS Institute, Cary, North Carolina, U.S.A.) was used.

RESULTS

Type of waste

Personal exposures to airborne dust, microorganisms and endotoxin during waste collection for all studies in this paper are presented in Tables 3, 4 and 5. Generally, the median concentrations of total microorganisms counted by microscopy were at levels of 10⁵–10⁶ cells m⁻³, whereas the concentrations of culturable fungi were 10⁴–10⁵ cfu m⁻³, and culturable bacteria 10³–10⁴ cfu m⁻³. Concentrations of Aspergillus fumigatus and actinomycetes were higher for garden waste (Table 5) compared with the other types of waste (Tables 3 and 4). Otherwise, the type of collected waste had only minor influence on the median concentrations of microorganisms.

Table 3. Concentrations of microorganisms, endotoxin and dust in air samples for workers collecting mixed household waste (study I, II, III) and sorted household waste in two-compartment containers (study IV). Median concentrations and ranges are given

<table>
<thead>
<tr>
<th>Truck</th>
<th>Study I basic</th>
<th>Study IIa basic</th>
<th>Study IIb exhaust</th>
<th>Study IIIa basic</th>
<th>Study IIIb basic</th>
<th>Study IVa high load</th>
</tr>
</thead>
<tbody>
<tr>
<td>Frequency of collection</td>
<td>7 days</td>
<td>7 days</td>
<td>7 days</td>
<td>7 days</td>
<td>14 days</td>
<td>14 days</td>
</tr>
<tr>
<td>N = 20</td>
<td>N = 6</td>
<td>N = 6</td>
<td>N = 6</td>
<td>N = 6</td>
<td>N = 6</td>
<td>N = 17</td>
</tr>
<tr>
<td>Total</td>
<td>180</td>
<td>1600</td>
<td>460</td>
<td>370</td>
<td>240</td>
<td>250</td>
</tr>
<tr>
<td>microorganisms cells ×10⁵ m⁻³</td>
<td>&lt;10–990</td>
<td>400–2100</td>
<td>&lt;10–1100</td>
<td>220–1700</td>
<td>95–260</td>
<td>97–1100</td>
</tr>
<tr>
<td>Fungi</td>
<td>74</td>
<td>240</td>
<td>38</td>
<td>75</td>
<td>100</td>
<td>15</td>
</tr>
<tr>
<td>cfu ×10³ m⁻³</td>
<td>11–490</td>
<td>26–340</td>
<td>23–57</td>
<td>&lt;0.1–470</td>
<td>68–810</td>
<td>4.3–200</td>
</tr>
<tr>
<td>Bacteria</td>
<td>5.0</td>
<td>20</td>
<td>3.2</td>
<td>24</td>
<td>1.6</td>
<td>1.2</td>
</tr>
<tr>
<td>cfu ×10² m⁻³</td>
<td>0.14–200</td>
<td>3.7–120</td>
<td>1.4–32</td>
<td>&lt;0.1–280</td>
<td>0.8–8.3</td>
<td>&lt;0.1–14</td>
</tr>
<tr>
<td>A. fumigatus</td>
<td>&lt;0.1</td>
<td>3.5</td>
<td>1.3</td>
<td>0.54</td>
<td>&lt;0.1</td>
<td>&lt;0.1</td>
</tr>
<tr>
<td>cfu ×10¹ m⁻³</td>
<td>&lt;0.1–2.4</td>
<td>&lt;0.1–2.10</td>
<td>&lt;0.1–9.1</td>
<td>&lt;0.1–1.9</td>
<td>&lt;0.1–5.2</td>
<td>&lt;0.1–46</td>
</tr>
<tr>
<td>Endotoxin</td>
<td>3.6</td>
<td>25</td>
<td>15</td>
<td>3.6</td>
<td>2.4</td>
<td>8.8</td>
</tr>
<tr>
<td>EU m⁻³</td>
<td>0–7</td>
<td>18–53</td>
<td>7–35</td>
<td>1.8–16</td>
<td>1.6–10</td>
<td>3.08–28</td>
</tr>
<tr>
<td>Dust</td>
<td>0.35</td>
<td>0.47</td>
<td>0.24</td>
<td>0.31</td>
<td>0.22</td>
<td>0.28</td>
</tr>
<tr>
<td>mg m⁻³</td>
<td>0.21–0.69</td>
<td>0.33–0.62</td>
<td>0.13–0.46</td>
<td>0.18–0.55</td>
<td>0.16–0.34</td>
<td>0.06–1.5</td>
</tr>
</tbody>
</table>

*Basic compactor truck (see text).
†Basic compactor truck with an exhaust system (see text).
‡Compactor truck with high loading (approximately 3 m above the ground).
Table 4 Concentrations of microorganisms, endotoxin and dust in air samples for workers collecting compostable household waste (study V and VI) in different seasons and using different truck designs.

<table>
<thead>
<tr>
<th>Truck</th>
<th>Study V a, b, c</th>
<th>Study VIa</th>
<th>Study VIb</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>compactor truck or platform truck</td>
<td>'bio-truck'</td>
<td>'Bates'</td>
</tr>
<tr>
<td>Season</td>
<td>Spring $N=23$</td>
<td>Summer $N=15$</td>
<td>Autumn $N=14$</td>
</tr>
<tr>
<td>Total microorganisms cells $x 10^3$ m$^-3$</td>
<td>240</td>
<td>&lt;10-1400</td>
<td>360-5800</td>
</tr>
<tr>
<td>Fungi cfu $x 10^3$ m$^-3$</td>
<td>46</td>
<td>360</td>
<td>660</td>
</tr>
<tr>
<td>Bacteria cfu $x 10^3$ m$^-3$</td>
<td>3.9</td>
<td>7.0</td>
<td>1.5</td>
</tr>
<tr>
<td>A. fumigatus cfu $x 10^3$ m$^-3$</td>
<td>0.38</td>
<td>1.2</td>
<td>&lt;0.1</td>
</tr>
<tr>
<td>Mesophilic actinomycetes cfu $x 10^3$ m$^-3$</td>
<td>—</td>
<td>0.91</td>
<td>0.72</td>
</tr>
<tr>
<td>Endotoxin EU m$^-3$</td>
<td>2.4</td>
<td>12</td>
<td>11</td>
</tr>
<tr>
<td>Dust mg m$^-3$</td>
<td>0.09-0.68</td>
<td>0.17-0.60</td>
<td>0.14-0.56</td>
</tr>
</tbody>
</table>

Data for the outdoor reference samples are presented in Table 6. The median concentrations of microorganisms and dust were always lower in the reference samples than in the personal samples.

For each sample, the concentrations of total microorganisms, culturable fungi, and culturable bacteria were classified into 'high', 'intermediate', and 'low'

Table 5 Concentrations of microorganisms, endotoxin and dust in air samples for workers collecting garden waste, recyclable waste, waste for incineration and paper (study IV, VII and VIII) in different seasons. Median concentrations and ranges are given

| Waste | Study VII garden | Study VIIla garden | Study VIIib 'bulky' Spring + Autumn $N=11$ | Study VIIlc recyclable Spring + Autumn $N=14$ | Study IVb paper Summer $N=10$
<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Season</td>
<td>Autumn $N=12$</td>
<td>Spring $N=6$</td>
<td>Autumn $N=11$</td>
<td>Autumn $N=11$</td>
<td>Summer $N=10$</td>
</tr>
<tr>
<td>Total microorganisms cells $x 10^3$ m$^-3$</td>
<td>590</td>
<td>320-1300</td>
<td>590</td>
<td>170-1200</td>
<td>500</td>
</tr>
<tr>
<td>Fungi cfu $x 10^3$ m$^-3$</td>
<td>160</td>
<td>12-450</td>
<td>370</td>
<td>77-660</td>
<td>250</td>
</tr>
<tr>
<td>Bacteria cfu $x 10^3$ m$^-3$</td>
<td>3.8</td>
<td>3.8</td>
<td>60</td>
<td>8.5</td>
<td>8.5</td>
</tr>
<tr>
<td>A. fumigatus cfu $x 10^3$ m$^-3$</td>
<td>6.2</td>
<td>0.8-15</td>
<td>4.1</td>
<td>17-280</td>
<td>5.5</td>
</tr>
<tr>
<td>Mesophilic actinomycetes cfu $x 10^3$ m$^-3$</td>
<td>2.0</td>
<td>2.2-27</td>
<td>—</td>
<td>0.48-50</td>
<td>4.5</td>
</tr>
<tr>
<td>Endotoxin EU m$^-3$</td>
<td>3.0</td>
<td>0.3-25</td>
<td>9.4</td>
<td>0.2-11</td>
<td>2.8</td>
</tr>
<tr>
<td>Dust mg m$^-3$</td>
<td>0.08-0.76</td>
<td>0.28</td>
<td>0.76</td>
<td>0.15-1.2</td>
<td>0.32</td>
</tr>
</tbody>
</table>
Table 6. Concentrations of microorganisms and dust in the outdoor reference air samples from all studies. Data are median concentrations with ranges in parenthesis. The median concentration of all reference samples and the 90% percentile of these are also given.

<table>
<thead>
<tr>
<th></th>
<th>Study I</th>
<th>Study II</th>
<th>Study III</th>
<th>Study IV</th>
<th>Study V</th>
<th>Study VI</th>
<th>Study VII</th>
<th>Study VIII</th>
<th>All studies median</th>
<th>All studies 90% percentile</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>N = 8</td>
<td>N = 3</td>
<td>N = 3</td>
<td>N = 8</td>
<td>N = 19</td>
<td>N = 3</td>
<td>N = 3</td>
<td>N = 16</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total microorganisms cells (\times 10^3 \text{ m}^{-3})</td>
<td>40</td>
<td>&lt; 10</td>
<td>150</td>
<td>33</td>
<td>21</td>
<td>&lt; 10</td>
<td>80</td>
<td>44</td>
<td>36</td>
<td>143</td>
</tr>
<tr>
<td>Fungi cfu (\times 10^3 \text{ m}^{-3})</td>
<td>24-47</td>
<td>&lt; 10-230</td>
<td>95-240</td>
<td>2.4-85</td>
<td>&lt; 10-600</td>
<td>&lt; 10-36</td>
<td>89-180</td>
<td>&lt; 10-210</td>
<td>0.52</td>
<td>3.5</td>
</tr>
<tr>
<td>Bacteria cfu (\times 10^3 \text{ m}^{-3})</td>
<td>1.4</td>
<td>0.53</td>
<td>0.68</td>
<td>0.75</td>
<td>0.80</td>
<td>0.37</td>
<td>0.37</td>
<td>0.24</td>
<td>&lt; 0.1</td>
<td></td>
</tr>
<tr>
<td>Dust mg m(^{-3})</td>
<td>&lt; 0.1-0.68</td>
<td>&lt; 0.1</td>
<td>&lt; 0.1</td>
<td>&lt; 0.1-3.4</td>
<td>&lt; 0.1-110</td>
<td>&lt; 0.1-15</td>
<td>&lt; 0.1-19</td>
<td>&lt; 0.1-0.38</td>
<td>0.04</td>
<td>0.10</td>
</tr>
</tbody>
</table>

Bacterial exposure in waste collection
concentrations. The limit for high concentrations was based on data from the literature (see discussion) indicating concentrations of microorganisms that might cause health problems. The limit for 'low' concentrations was defined as the 90% - percentile for all reference samples (Table 6). Following these criteria, the level of total microorganisms was considered to be 'high' if the concentration was more than $10^6$ cells m$^{-3}$ and low when less than $1.6 \times 10^5$ cells m$^{-3}$ of air. The limits for culturable fungi, bacteria and *Aspergillus fumigatus* considered 'high' were set to $10^5$ cfu m$^{-3}$, $10^4$ cfu m$^{-3}$ and $10^4$ cfu m$^{-3}$, respectively. Likewise, the concentrations were considered 'low' when less than $2.2 \times 10^3$ cfu m$^{-3}$, $3.6 \times 10^2$ cfu m$^{-3}$ and $10^2$ cfu m$^{-3}$ for fungi, bacteria and *A. fumigatus* (the latter was set to the detection limit as the reference samples were not analysed for *A. fumigatus*). Figure 1 show the percentage of samples with 'high' and 'low' levels of total microorganisms, fungi, bacteria, and *A. fumigatus* for the main types of waste. Total counts of microorganisms [Fig. 1(a)] were generally low: 12–17% for garden, sorted, mixed and compostable waste. The proportion of samples with 'high' concentrations of bacteria [Fig. 1(c)] was fairly large for most types of waste, highest for garden waste (45%), the paper/glass fraction (43%), and mixed waste (38%). For garden waste,
76% and 41% of samples were high with respect to culturable fungi [Fig. 1(b)] and *A. fumigatus* [Fig. 1(d)], respectively. Mixed and compostable waste had 38% and 23% of samples in the high category of fungi. Otherwise, less than 20% of samples were high with respect to fungi and *A. fumigatus*.

The median concentration of thermophilic actinomycetes was 150 cfu m\(^{-3}\) in samples from collection of garden waste. Otherwise thermophilic actinomycetes were found in less than 50% of the samples from the other types of waste, but concentrations up to 2–5×10\(^4\) cfu m\(^{-3}\) were occasionally detected in samples from workers collecting garden waste, compostable household waste and recyclable paper/glass (data not shown).

**Equipment**

Compostable household waste was collected using a total of five different combinations of collection systems in study V (compactor truck/container, compactor truck/sack, platform truck/sack) and study VI (‘bio-truck’/container, ‘bio-truck’/sack). One of the four sampling periods of study V was in the same season as the only sampling period of study VI (late spring/early summer 1994). Exposure data obtained in this period are compared in Fig. 2. The three equipment combinations with the compactor truck and platform truck resulted in similar exposure levels: fungal concentrations ranged from 3×10\(^4\) to 8×10\(^4\) cfu m\(^{-3}\) and bacterial concentrations from 3×10\(^3\) to 7×10\(^3\) cfu m\(^{-3}\). The use of a biotruck (high loading of the waste) to collect household waste in containers decreased the exposure level compared to the common compactor truck (low loading of the

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**Fig. 2.** Median exposure to fungi and bacteria for persons collecting compostable household waste using compactor truck, platform truck (study V) or bio-truck (study VI). The waste was kept in containers or sacks at the households. Only data from the same season are included (late spring/early summer).
waste): the concentration of fungi was lower by a factor of 25 ($P = 0.003$) compared with the common compactor truck and the bacterial concentration was lower by a factor of 7 ($P = 0.10$). The use of the 'bio-truck' designed specifically for disposal of paper sacks ('Bates system') gave just as high concentration of fungi as the compactor truck, but significantly lower concentration of bacteria ($P = 0.01$), which was also lower than the bacterial concentrations by any other system used for collection of compostable waste. However, the concentration of dust was very low for both 'bio-trucks' (medians ≤0.15 mg m$^{-3}$). Therefore, the median concentration of fungi per gram of dust was 4–7 times higher for the 'bio-truck'/sack system compared to compactor and platform trucks and higher by a factor of 46 compared to the 'bio-truck'/container system. For all systems, the concentration of total microorganisms determined by microscopy was 2–6 times higher than the concentration of culturable fungi.

Different trucks and equipment were also used for collection of mixed household waste (weekly collection of waste) and personal air samples were collected in the summer time of 1993 and 1994 (Table 3). Compactor trucks and different sizes of two- and four-wheel containers and bins were used (Table 1). In these studies, the median concentration of fungi was highest ($2.4 \times 10^5$ cfu m$^{-3}$) for the basic compactor truck and containers (study IIa), and ranged from $4 \times 10^4$ to $8 \times 10^4$ cfu m$^{-3}$ in the other studies. Study II was designed to assess the occupational exposure during collection of mixed household waste in containers emptied into a compactor truck improved with an exhaust system as compared with the basic compactor truck. Clearly, the truck with exhaust ventilation resulted in lower exposure levels to microorganisms in relation to the basic truck in study II. However, this level cannot be considered low when compared with the other studies, as the basic compactor truck/containers in study I resulted in similar concentrations of microorganisms. Sorted household waste kept in two-compartment containers was collected using compactor trucks with high loading well above the breathing zone of the workers. The exposure to fungi and bacteria was fairly low compared to collection of mixed household waste using the basic compactor truck.

**Seasonal variation**

Within studies V and VIII, bioaerosol measurements were obtained in the same districts (same type of waste, equipment, etc.) in different seasons. Measurements on collection of garden waste, bulky waste for incineration, and recyclable materials were done in spring and autumn (similar air temperatures). In these periods, there were only minor differences in the concentrations of bioaerosols (Table 5). Measurements on collection of compostable household waste in study V covered all four seasons (Table 4). The season had statistical significant influence ($P < 0.001$) on the concentration of total microorganisms, culturable fungi, *A. fumigatus*, and endotoxin with the lowest concentrations in the winter. The bacterial concentrations showed no clear seasonal variation.

**Indicators for screening of microbial exposure**

For all personal samples, the concentration of total microorganisms counted by microscopy, culturable fungi and culturable bacteria were plotted against the concentration of dust [Fig. 3(a–c)], and the concentration of bacteria was plotted
against the concentration of endotoxin [Fig. 3(d)]. The concentration of culturable fungi or culturable bacteria were plotted against the concentration of total microorganisms [Fig. 4(a–b)]. These scatter plots indicated that there was only a weak relationship between the microbial parameters and the dust concentration, and a slightly better relation between the culturable microorganisms and total counts of microorganisms. There seemed to be no relation between the concentration of bacteria and endotoxin. Four-fold table analysis was performed (Table 7) for dust as an indicator of microbial exposure and for total microorganisms as an indicator of exposure to viable fungi or bacteria. The screening technique had a rather high sensitivity (Se = 0.66–0.88) and a high predictive performance for negative screening test results (P_{v2} = 0.82–0.95). However, the performance for positive screening test results was poor (P_{v1} = 0.20–0.46).

DISCUSSION

Workers are exposed to bioaerosols while collecting household waste and further down-stream in the waste treatment process. The incidence of work-related pulmonary, gastrointestinal and skin problems may be greater in waste collectors than in the general work force (Poulsen et al., 1995a). Knowledge of the aetiology of these problems is limited, but it is likely that they are caused by exposure to bioaerosols or volatile organic compounds. Studies in other occupational environments with high exposure to bioaerosols, have shown that exposure to bioaerosols may cause a wide range of respiratory and mucosal symptoms that vary in severity from mucous membrane irritation to acute or chronic diseases such as toxic alveolitis (also called organic dust toxic syndrome, ODTS), allergic alveolitis and asthma (reviewed by Lacey and Dutkiewicz, 1994). However, it is far from resolved which components in bioaerosols are responsible for the effects on the respiratory system, but it seems that inhalation of fungal and actinomycete spores, gram-positive and gram-negative bacteria, endotoxin from gram-negative bacteria and probably glucans from the fungal cell wall are all likely to be implicated in the causes of respiratory symptoms. In a few occupational environments, it has been shown that there is a dose–response relationship for a certain bioaerosol component. This is particularly the case in the cotton industry, where it was found that the exposure to endotoxin relates to decreased lung function, and the 'no effect level' for endotoxin in cotton dust was found to be 10 ng m^{-3} (Castellan et al., 1987) or 33 ng m^{-3} (Rylander et al., 1985). An exposure–response relationship for endotoxin was also found for workers in wastewater treatment plants (Laitinen et al., 1992) and swine confinement buildings (Donham et al., 1989; Heederick et al., 1991). In Danish waste handling plants (sorting of household waste), Sigsgaard et al. (1994) could find no clear relationship between symptoms and measurements of dust, endotoxin or microorganisms, but compared with control groups, waste-handling workers had significantly higher prevalence of upper respiratory tract symptoms and eye problems. For these garbage handling workers the mean exposure was fairly low: $4.6 \times 10^4$ cfu m^{-3} total viable microorganisms, $1.4 \times 10^4$ cfu m^{-3} fungi, and $4.8 \times 10^3$ cfu m^{-3} gram-negative bacteria. Eduard et al. (1994) reported that for sawmill workers, exposure levels exceeding $10^6$ fungal spores per m^{3} air were related to increased prevalence of respiratory symptoms, mucous membrane irritation and
Fig. 3. Concentration of microbial parameters plotted against the dust concentration or endotoxin concentration. Each data point ($N = 196$) represents one sample, and personal samples from all studies are included. (a) Total microorganisms vs dust; (b) fungi vs dust; (c) bacteria vs dust; (d) Bacteria vs endotoxin. Dashed lines represent the limit for 'high' concentration and the screening limit presented in Table 7. Values below the limit of detection are marked at half the limit of detection.
Fig. 4. Concentration of (a) fungi and (b) bacteria against the concentration of total microorganisms. Dashed lines represent the limit for 'high' concentration and the screening limit presented in Table 7. Values below the limit of detection are marked at half the limit of detection.

ODTS-like symptoms. In that study, fungal spores were quantified by electron microscopy, that is, including culturable and non-culturable spores analogous to the counts of total microorganisms in the present study. Malmberg et al. (1988) reported that long term exposure above $10^8$–$10^9$ fungal spores per m$^3$ air may cause allergic
alveolitis. For other bioaerosol components and other occupational environments, information on dose–response relationships is scarce, and none exists for waste handling (see Millner et al., 1994). Therefore, on the basis of the existing information, it is impossible to set limits for 'safe' and 'hazardous' exposures for any of the bioaerosol components that are found in connection to waste collection. However, unofficial occupational exposure limits (OEL) have been suggested for some bioaerosol components by researchers or by national authorities. Recently Malmros (1990) suggested a limit for acceptable air quality of 100 ng m\(^{-3}\) for endotoxin, 10\(^3\) cfu m\(^{-3}\) for gram-negative bacteria and 10\(^4\) cfu m\(^{-3}\) for total bacteria. In Poland an OEL for total viable microorganisms has been suggested at the level of 10\(^5\) cfu m\(^{-3}\) (Dutkiewicz, 1992). A Dutch working group on indoor air pollution reported that bacterial and fungal concentrations above 10\(^4\) cfu m\(^{-3}\) in total or above 500 cfu m\(^{-3}\) for each specific group of organisms or species of a potentially pathogenic nature, should be considered a threat to the worker's health (Dutch Occupational Health Association, 1989 cited from Heida et al., 1995). It should be noted that the outdoor concentration of airborne fungal spores are usually in the order of 10\(^3\)-10\(^4\) m\(^{-3}\), changing with weather, season, geographical location, and so on (Lacey and Crook, 1988).

For the present study a three-way classification ('high', 'intermediate', and 'low') of exposure to microorganisms was used for the graphical presentation of data. It has to be emphasized that the criteria for the classification was a clear presentation of data and not to suggest any occupational exposure limits.

While the median concentrations of airborne microorganisms showed only small differences between collection of different types of waste, the classification of each sample into 'high' and 'low' concentrations made it possible to differentiate clearly the exposure levels on the basis of type of waste. In addition, the median exposure in a given job is probably not the most important value with respect to the risk of developing health problems. The probability of having working days with a high exposure may be a more valuable measure. The classification showed that the collection of garden waste leads to a high percentage of the samples in the 'high' category for mesophilic fungi (76%) and the thermophilic fungus A. fumigatus (41%), while collection of 'bulky waste' and paper resulted in no samples with 'high' concentrations of these fungal groups. Though only few samples had 'high' levels of total microorganisms, none of the samples from collection of garden waste had 'low'

### Table 7. Result of the four-fold table classification together with the screening limit (SL) for which the classifications were calculated (169 observations)

<table>
<thead>
<tr>
<th>Microbial parameter</th>
<th>Sp</th>
<th>Se</th>
<th>Va</th>
<th>Pv₁</th>
<th>Pv₂</th>
<th>SL</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dust as an indicator of exposure to the microbial parameter</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total microorganisms</td>
<td>0.57</td>
<td>0.79</td>
<td>1.36</td>
<td>0.20</td>
<td>0.95</td>
<td>0.30</td>
</tr>
<tr>
<td>Fungi</td>
<td>0.65</td>
<td>0.66</td>
<td>1.31</td>
<td>0.44</td>
<td>0.82</td>
<td>0.31</td>
</tr>
<tr>
<td>Bacteria</td>
<td>0.66</td>
<td>0.69</td>
<td>1.35</td>
<td>0.44</td>
<td>0.85</td>
<td>0.31</td>
</tr>
<tr>
<td>Total microorganisms as an indicator of exposure to the microbial parameter</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fungi</td>
<td>0.57</td>
<td>0.88</td>
<td>1.45</td>
<td>0.46</td>
<td>0.92</td>
<td>2.5×10(^5)</td>
</tr>
<tr>
<td>Bacteria</td>
<td>0.47</td>
<td>0.78</td>
<td>1.25</td>
<td>0.36</td>
<td>0.85</td>
<td>2.3×10(^5)</td>
</tr>
</tbody>
</table>
Bioaerosol exposure in waste collection

Concentrations (<1.6×10^5 cells m⁻³). Compostable, mixed and sorted household waste had less samples in the 'high' category with respect to all parameters as compared to garden waste. The recyclable fraction had a high percentage of samples with 'high' level of bacteria, and collection of this fraction was the only type of waste that resulted in almost even median exposure to bacteria and fungi. For all types of waste, concentrations of endotoxin in the samples ranged from 0 to 100 EU m⁻³ which was lower than the 'no effect level' of 10 ng m⁻³ (equivalent to 120–150 EU m⁻³) reported by Castellan et al. (1987). The concentrations of dust were also generally low, and all samples were less than the Danish occupational exposure limit for organic dust (3 mg m⁻³).

Several factors other than type of waste may govern waste collectors' bioaerosol exposure, for example type of equipment for storage of the waste prior to collection, type of truck used for collecting the waste, and the weather conditions. The different types of truck used for collection of mixed and compostable household waste seemed to influence the exposure level. The use of 'bio-trucks' for collection of compostable household waste caused lower exposure to dust and bacteria compared to the common compactor truck. This is most likely because loading the 'bio-trucks' generates less bioaerosols as the waste is not compressed. In addition, the workers are farther away from the source of exposure as the 'bio-trucks' are loaded from the top (approximately 3 m above the ground), whereas loading of the common compactor truck takes place close to the worker's breathing zone. In a study on collection of household waste with a compactor truck, it was found that the concentration of microorganisms was noticeable lower 2–3 m behind the truck (<10^3 cfu m⁻³) as compared with close to the scoop of the truck (10^4–10^5 cfu m⁻³) (Ducel et al., 1976).

Recently we have used a rotating drum tester to characterize dustiness of compostable waste and found that the microbial content of the dust (potency of the dust) was affected by storage conditions of the waste prior to collection (Breum et al., 1997). For waste kept in paper sacks the potency of the dust was higher by at least a factor of 30 for fungi compared to waste kept in containers. In the present study, the potency of the dust sampled at the biotruck/sack system was higher by a factor of 46 compared with that sampled at the biotruck/container system. The growth of organisms in the waste and thereby also the potential for bioaerosol generation from a given type of waste might also be influenced by the weather conditions in which the waste has been kept. In study V workers' exposure to bioaerosols was measured during the collection of compostable household waste throughout all four seasons of the year, and the lowest exposure was found to be in winter time. A seasonal variation in exposure to bioaerosols was also seen for workers at waste composting plants (Hirn et al., 1982), waste recovery plants (Lembke and Kniseley, 1985) and waste transfer stations (Crook et al., 1987).

As mentioned above, the exposure to dust during collection of waste was fairly low, but as the dust analysis is less costly and time consuming than the analysis of any of the microbial parameters, it would be of great value if dust measurements could be used to predict the level of the other bioaerosol parameters, that is, if dust exposure could be used as an indicator of microbial exposure. Alternatively, microscopy counts of total microorganisms would be a convenient indicator as the samples can be stored frozen before analysis and the analysis is less expensive
compared with culturing on a number of different media. By using the abovementioned limits for 'high' concentrations of the microbial parameters, the effectiveness of using dust or total microorganisms as indicators was tested on the present bioaerosol data. Diagnostic methods are often described in terms of the sensitivity, the specificity and the validity. Maximizing the validity (Va) may be useful for establishing the screening limit of a screening test. The value of Va should approach 2 as nearly as possible, but when it is important to detect hazardous situations the sensitivity (Se) should approach 1 at the cost of the specificity (Zielhuis and Verberk, 1974). For this study the screening limit was derived from the criteria of maximizing the validity and keeping the sensitivity higher than the specificity. For practical applications it is important to know the performance of the screening test to predict the concentration of the true microbial parameter. For concentrations of the indicator below the screening limit the test would correctly classify the concentration of the true parameter as being 'low' or 'intermediate' for 82–95% (PV2) of the samples (Table 7). For situations with concentrations of the indicator above the screening limit the test had a rather poor performance with only 20–46% (PV1) of the samples having 'high' concentrations of the true parameter. However, if such large misclassification is unacceptable the situation can be improved by analysing all samples exceeding the screening limit for content of the true parameter.

It was clear from Fig. 3(d) that bacterial concentration related poorly to endotoxin, and no further analysis was made on the performance of endotoxin as an indicator. In the air of wastewater treatment plants, a good correlation was found between endotoxin and gram-negative bacteria and a fairly good correlation between endotoxin and total viable bacteria (Laitinen et al., 1992). Zock et al. (1995) also reported a good correlation between the concentration of endotoxin and gram-negative bacteria in the Dutch potato processing industry. The poor relationship between endotoxin and total bacteria in the air of waste collectors is probably explained by the fact that gram-negative bacteria only constitute a minority of the total culturable bacterial flora (Nielsen et al., 1995a).

Though the screening test using the dust concentration as indicator performed in an acceptable manner as tested with the data in this study, it might not be the case with data sampled in other occupational environments. Certainly, if the screening test should be used in other environments or even for waste collectors with different working conditions, the screening value would have to be redefined to obtain maximal validity of the test. It is more likely that counts of total microorganisms is a robust indicator for culturable counts of bacteria and fungi as these, of course, are included in the microscopy count. However, the relative number of fungal spores, bacterial cells, actinomycetes spores, and the fraction of culturable organisms would be expected to vary significantly in different occupational environments. For the present data, the best indicator was total microorganisms as indicator of culturable fungi (Va = 1.4). This was to be expected as the ratio of fungi to bacteria generally was about 10, meaning that most of the microorganisms counted by microscopy would be fungal spores.

In several of the studies (studies I, IV, V, VIII) we have also evaluated the health status of the waste collectors by clinical examination, questionnaires, peak flow measurements, and analysis of blood samples. Generally the waste collectors in the
clinical examinations were healthy, and no work related effects were found, except in
the group collecting garden waste in plastic sacks (study VIII). The symptoms
reported by these workers included airway irritation and asthmatic symptoms which
was probably related to the special way of handling the waste (manual emptying of
sacks into the scoop of the truck) (Stenbæk et al., 1996).

Furthermore, we are currently linking the exposure data reported in the present
study to data from a nation-wide questionnaire based survey among waste
collectors. The preliminary analyses indicate that waste collectors using equipment
or work routines resulting in high levels of bioaerosol exposure have comparatively
high prevalences of bronchitis and gastrointestinal complaints (Hansen et al., 1997;
Ivens et al., 1997).

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