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Bedrooms without house dust mites

Final report from a preliminary study.



Lars Wadsö
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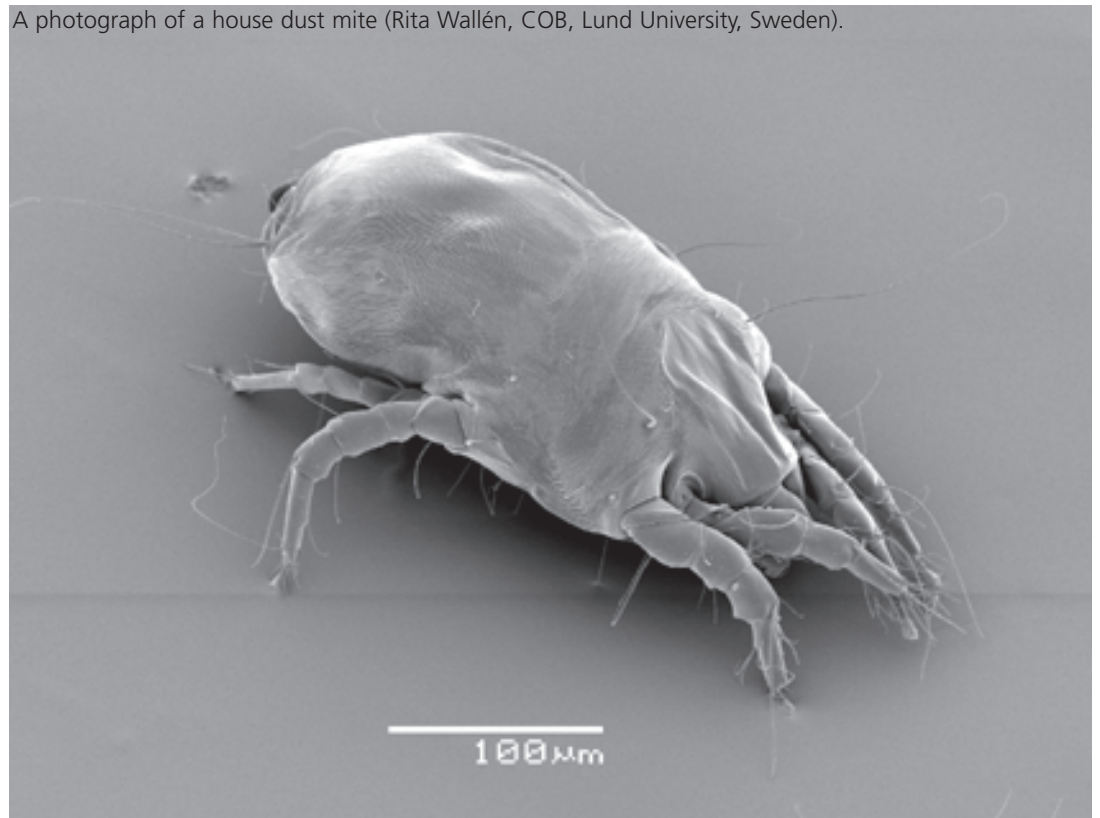
Report TVBM-7181, TVBH-7230

Lund 2005

Bedrooms without house dust mites

Final report from a preliminary study.

A photograph of a house dust mite (Rita Wallén, COB, Lund University, Sweden).



Lars Wadsö
Kaisa Svennberg

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Summary

During the last decades there has been a rapid increase in allergies in Sweden and in other industrialized countries and house dust mites (HDM) allergens are among those that cause most problems. HDM are small spider animals that are commonly found in beds where they survive on skin scales. Several studies have shown that the concentration of house dust mites is lower in dry environments. This has also been studied experimentally and it is generally believed that house dust mites cannot live at relative humidities below 50%. Studies also show that allergic persons have less symptoms during the dry winter period.

House dust mites are uniquely fitted for a life in dry environments and can without problems survive long periods at low humidity, as long as there are daily periods of higher humidity. The reason for this is that the mites have means for extracting moisture directly from the air. A hygroscopic salt solution flows in a furrow on the outside of their bodies. When it enters their bodies again it has absorbed water that is extracted and used to replenish their water reserves. This mechanism works down to about 58% RH at 20°C. Another adaptation to dry environments is that several of HDM developmental stages are very desiccation tolerant, for example can their eggs survive long dry periods to later hatch when the humidity is higher.

What is mentioned above makes it difficult to eradicate house dust mites simply by lowering the relative humidity. At 20°C one must go down to 58% RH the whole year in all places where the mites stay. However, the mites are sensitive to higher temperatures. At increased temperatures will their moisture extraction mechanism only work at higher RH and after 40 minutes at 45°C they will die. It therefore seems that combining lowering of the humidity level with increased temperature is a possible mean of controlling house dust mites.

One other factor that makes it difficult to get rid of HDM is that they move quite rapidly, possible sensing in which directions they can find the optimal conditions. They can probably move around in most mattresses.

Several studies show that the concentration of HDM is lower in winter when it is drier indoors. Apart from this, investigations on HDM and their allergens show mixed results. Only a few studies have shown that a general drying of the indoor environment is beneficial. One reason for this is that one normally does not measure or control the humidity in the environment where the HDM live, both only the room RH. Laundry, freezing and treatment with steam has been shown to be efficient ways of decreasing the number of living HDM. It is essential that the whole bed is treated. Mattress encasings (semi-permeable covers of the mattress) does not seem to be as efficient as one could expect.

Several researchers have realized that to get rid of HDM in the environment where they live, for example in a bed, one must study the microclimate and how they can survive it. One conclusion of their (and our) investigations is that a sleeping person produces water, but that the temperature near the body is so high so that the microclimate is unsuitable for HDM. It is therefore probable that the HDM move in the beds to the places with the best microclimates. However, there are today no useful models for how they move in response to humidity and temperature.

We have tested experimental methods to study the activity of HDM as a function of humidity and temperature. In these experiments a laboratory culture of *Dermatophagoides farinae* (Df) was used. The main method was isothermal calorimetry, i.e. the measurement of the heat produced by the mite respiration. The method worked well, but the activity of the animals generally decreased after about one month; probably because the food supply did not last longer. Studies with different humidity levels showed an expected effect and a study of the temperature sensitivity successfully showed that Df die after 40 minutes exposure to 45°C. Isothermal calorimetry is a both efficient and simple method to quantify HDM activity.

We also used an visual method to count living HDM on Petri dishes placed in different environments. This study also gave an expected result. A separate study was also made in which the HDM were automatically counted by photography and image analysis. It was possible to simultaneously determine the number of moving HDM and their sizes and velocity. It is possible to use this method in the future to study how HDM move in response to humidity and temperature gradients.

We have developed a measurement system for temperature and relative humidity that can be used in beds. We use small capacitive RH-sensors and thermistors and the system has worked well under test measurements in two beds. Our measurements show that the temperature increases significantly under a sleeping person, but the relative humidity is quite the same as during the day. It is therefore not possible for the mites to make use of the humidity in the top part of the bed while the person is still in the bed. We could also see effects of moisture buffering, for example that the RH increased during the day in some parts of a bed.

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1. Introduction

The last decades have seen a rapid increase in allergies in Sweden and many other countries. Although allergenic sensitivity to cat, dog and different types of pollen are common, the most common such reactivity in, e.g., Southern Sweden and Denmark, is that to house dust mites (HDM). These small, hardly visible animals are well adapted to survival in the human indoor habitat; for example in beds, sofas and carpets, where the relative humidity (RH), at least a few hours per day, can reach quite high levels.

Previous studies show that in drier environments the house dust mite occurrence is lower, and that persons with dust mite allergy have fewer symptoms during the winter when the indoor relative humidity is low. There have also been attempts to alter the microclimatic conditions in beds to reduce the house dust mite occurrence. The microclimatic conditions in beds have been studied both in field measurements and by modeling but there is still a lack of knowledge of how the hygrothermal material properties of the mattress and bedding effect the environmental conditions for the house dust mites.

This is the final report from the preliminary study Bedrooms Without House Dust Mites funded by SBUF - the Development Fund of the Swedish Construction Industry. We acknowledge the help of the other persons in the informal Multi-disciplinary House Dust Mite Project at Lund University who have helped and encouraged us in this project: Morgan Andersson (Otorhinolaryngology, Lund University), Christer Hansson (Dermatology and Venereology, Lund University), Lars-Erik Harderup (Building Physics, Lund University), Lars R. Lundqvist (Zoological Museum, Lund University). We also thank Kajsa Mellbrand who helped us with the mite measurements and Måns Österberg (Allergon AB) for the generous gift of living house dust mites. It is our intention to continue this project with a multi-disciplinary project aiming to find technical solutions for reduction of house dust mites in bedrooms by environmental control.

2. Literature survey

Our bedrooms are a habitat that house dust mites (HDM) have been able to occupy very successfully by an efficient water management strategy. All organisms must have strategies to deal with water, as no life as we know it, is possible without water as all biochemical processes take place in an aqueous environment. Many organisms have developed advanced strategies to find water or to minimize the loss of water. Desert cacti can withstand long periods of drought, as they can store large amounts of water and effectively change the volume of their bodies when they lose part of the stored water (Mauseth, 2000). Cockroaches can sense gradients in relative humidity, and thus escape from too dry or too moist environments (Tichy, 2003). So called resurrection plants can survive total dehydration and return to fully functional photosynthesis within 48 h of being exposed to water again (Scott, 2004). Also HDM have developed interesting humidity-oriented strategies for survival in dry environments such as our houses.

This review concerns HDM survival in bedrooms and beds. It starts with a review of HDM and their survival at different climatic conditions; then follows a review of what is known about HDM and microclimate in bedrooms. The nomenclature used in this review is given below.

a_w	Water activity
CEH	Critical Equilibrium Humidity (lowest constant RH at which HDM survive). The term CEA (Critical Equilibrium water-vapor Activity) is sometimes used instead of CEH (de Boer, 1990).
<i>Dp</i>	<i>Dermatophagoides pteronyssinus</i> (“European HDM”)
<i>Df</i>	<i>Dermatophagoides farinae</i> (“American HDM”)
HDM	House Dust Mite
RH	Relative Humidity

The relative humidity (RH) is the vapor pressure of a gas divided by the saturation vapor pressure (the maximal vapor pressure at the temperature in question). It is usually expressed in percent, e.g. at an RH of 50% half the capacity of taking up water is used. The water activity (a_w) is similar to RH in its definition, but is also used for the humidity state of solids and liquids. It also has a more rigorous thermodynamic definition, but for our purposes it has the same value as the RH at equilibrium. A passive object, e.g. a piece of wood, will have a water activity of 0.50 if it is in equilibrium with air at 50% RH. Water has a water activity of 1.00.

There has been a discussion if critical humidity levels should be represented as RH or as absolute vapor content (g water vapor per m³ or kg of air, essentially proportional to vapor pressure). Most works seems to prefer RH, but some, e.g. Hart, (1998), discuss HDM water relations in terms of vapor content. As an example, Hart (1998) writes “Larger house-dust mite populations are found when the absolute indoor air humidity is above 7 g/kg (45% relative humidity at 20 °C)”. We believe that both these humidity concepts are useful, as some

processes (sorption, metabolic processes) are governed by RH, while diffusion processes are governed by absolute humidity. HDM population growth cannot be modeled only by one of these parameters. Two parameters are needed, e.g. RH and temperature (which then uniquely also defines the absolute humidity); see also discussions in Cunningham (1996); and Toolson (1980).

House dust mites

Mites

Mites are a very diverse and wide-spread group of animals that can be found in almost any habitat on earth. In the words of Schauff (2000), they are “ubiquitous, inconspicuous, harmful and helpful”. More than 30 000 species have been identified, some feeding on plants or fungi, while others, for example bird mites, have developed complex parasitic relationships with other animals.

The mites we are mainly concerned with here, are called house dust mites (HDM), but there are also other types of mites, for example storage mites, that can live indoors. Two HDM species, *Dermatophagoides pteronyssinus* (*Dp*, “European HDM”) and *D. farinae* (*Df*, “American HDM”), that are most common in our homes. The common names are unfortunate as both are found both in Europe, America and in other places (Arlan, 2001). These two species are often found in mixed populations. A third house dust mite *Euroglyphus maynei* is less frequent and more limited in geographical distribution than *Dp* and *Df* (Arlan, 2001) and is also less desiccation tolerant (Arlan et al., 1998).

Another group of mites that we come in contact with are the storage mites. These are frequently found in grain, hay, dried foodstuffs etc., and are also sources of allergens (Hart, 1990; Vidal et al., 2004). Storage mite sensitivity is of major importance in rhinitis and asthma in farmers (Arlan, 2001). Two common storage mites are *Lepidoglyphus destructor* and *Tyrophagus putrescentiae*. Swedish homes mainly contain *Dp*, *Df* and the two storage mites mentioned above (Warner et al., 1999).

We may also come in contact with the mite *Sarcoptes scabiei* that colonizes the corneous layer of the human skin causing scabies. This mite is a true parasite and cannot live in any other conditions (Fain, 1990). There is also a large group of mites that normally parasitize birds, mice, rats etc. and that may bite humans when they do not find their preferred hosts (Arlan, 2001).

In biology, species (essentially a group of organisms that can reproduce) are ordered in a Linnean binomenclature system. For example is *Picea abies*, the spruce tree common in Sweden, of the *abies* species in the genus *Picea*. In this hierarchical system, related genera belong to the same family, related families belong to the same order, related orders belong to the same class etc. According to modern classification of mites, *Df* and *Dp* are related to *Euroglyphus*, being in the same family *Pyroglyphidae* in the order *Astigmata*. Of the same order, but in the families *Acaridae* and *Glycophagidae*, we find the storage mites *Tyrophagus putrescentiae* and *Lepidoglyphus destructor*. At the same distance from HDM in the classification scheme is also *Sarcoptes scabiei* in the family *Sarcoptidae*. However, predatory mites like *Gamasina* and *Cheyletiella* (that is a possible predator on HDM) belong to the order *Mesostigmata* and *Prostigmata* respectively, and are thus not as closely related to HDM as are the storage mites and *Sarcoptes*.

All mites and ticks belong to the sub-class *Acari* (spiders belong to the sub-class *Arachnides*) and the term “acari” is generally used for such animals. Thus, an acaricide is a substance that is toxic to ticks and mites, and *Experimental and Applied Acarology* is one of the most popular scientific journals dealing with ticks and mites. HDM and related mites are often called “pyroglyphid mites” (or “the pyroglyphidae”) after the name of the family to which they belong.

HDM anatomy and feeding

HDM are classified as invertebrates, as they do not have an internal skeleton. Like spiders, mites have four pairs of legs, but unlike spiders their body is undivided (true spiders have two body parts; insects have three pairs of legs and three body parts). HDM are free-living, but have many anatomical features in common with parasitic mites (Hart, 1990), from which they may have evolved.

Figs. 1-3 show a photograph and schematic drawings of HDM. They have a size of about 500 μm and a mass of 5-10 μg (Arlan and Wharton, 1974). Their bodies (*idosoma*) are divided into two parts by a transverse furrow (*sejugal furrow*), (the main part of the following morphological description is taken from Fain, 1990). They have no true head, but bear their mouthparts on the front (*anterior*) part of their body. The mouth parts consist of movable sensorial *pedipalps* and fang-like *chelicerae*. Mites have no antennae. There are four pairs of legs (except the larva, which only have three pairs) on the lower (*ventral*) side (the upper side is called *dorsal*). Each leg is formed of six segments ending with a claw. Mites have body hairs (*setae*) that are important for determination of species, e.g., HDM have short setae compared to storage mites, which are distinctly hairy. The rear (*posterior*) part contains the anus.

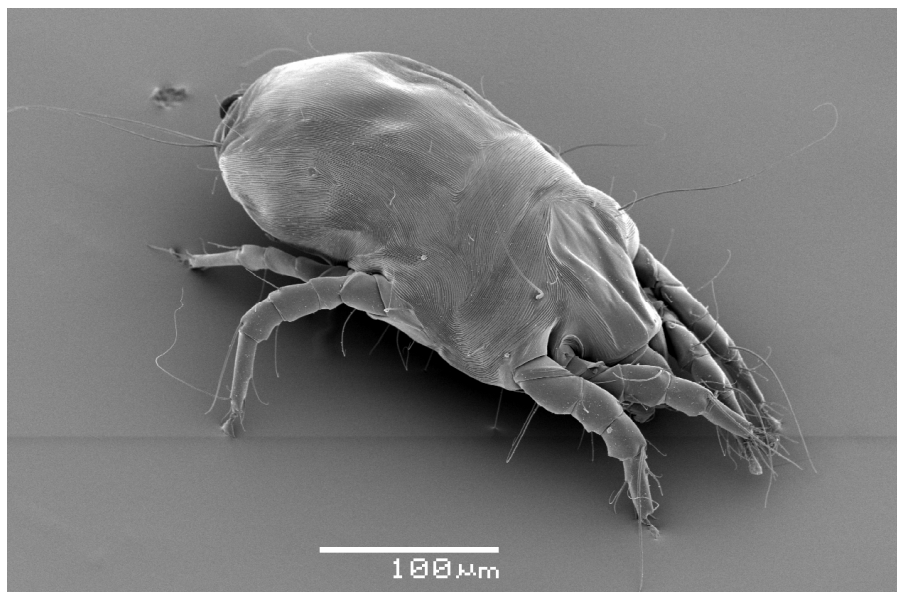


Figure 1. A photograph of a house dust mite (Rita Wallén, COB, Lund University, Sweden).

Mites have an exoskeleton (an outer skeleton, a shell), called the cuticle, just like spiders, insects and crustaceans. In the HDM, the dorsal part of the cuticle is striated in different ways for different species, a feature that can be used in the identification (Figs. 2 and 3). The HDM cuticle is soft, enabling the body to change size. This is used both when walking, in which the

whole body is deformed, as well as during desiccation, when the body deflates when water is lost (see Figs. 2 and 3 in de Boer et al., (1998) for an example). Other mites are often more sclerotized.

The mite cuticle contains glands that open onto its surface (Hart, 1990). Most important for the present review are the pair of supracoxal glands situated behind the front legs (Wharton and Furumizo, 1977). They secrete hygroscopic solutions into the associated podocephalic canal that runs towards the mouth parts, and is used for the uptake of water from the air (see section on Active uptake of water from unsaturated air p. 16).

Inside their bodies, haemolymph (body fluid) surrounds their internal organs (Hart, 1990). From the mouth parts leads a gut to excretory tubes and the anus. Their digestive system produces spherical faecal pellets that are approx. 20 μm in diameter (they do not urinate (Spieksma, 1997)). Unlike most terrestrial insects and other mites, HDM lack an organized respiratory system and associated openings (Arlian, 1992). Their oxygen uptake is by passive diffusion through the cuticle into the haemolymph. Oxygen consumption rates of 0.008 $\mu\text{g}/\text{h}$ has been measured on *Df* females and active and quiescent protonymphs consume 0.11 and 0.003 $\mu\text{g}/\text{h}$ (Hart, 1990).

HDM are generally believed to digest human skin scales (Hart, 1990; Spieksma, 1997), but they can also survive on a number of other nutritional sources. They do not drink (Spieksma, 1997). It seems to be essential that their food has a high mineral, fat and protein content. It has been reported that mites often are associated with moulds and that they may even need the moulds to pre-digest the skin scales or to provide vitamins or other essentials (Hart, 1990). However, although HDM certainly can eat mould hyphae and spores (as do other types of mites), the association between moulds and mites seems to be of secondary importance as HDM grow well at lower RH, for example 70%, than moulds can do. A review of this subject is given by van Asselt (1999).

Identification of mite species

Fain (1990) gives detailed morphological descriptions of mites causing allergy in man, that can be used in identification. For the mite species found indoors in Sweden, the storage mites are distinctly more hairy than HDM. The latter have the same number of setae, but these are significantly shorter. The setae stick out like whiskers from the legs and from the dorsal part of the body (Figs. 1-3). For the two HDM species *Df* and *Dp* that we are mainly concerned with here, the females are distinguished by the different dorsal striations (Figs. 2-3) and that *Df* has a broader body. The *Df* male differs from the *Dp* male in that the shape of the posterior dorsal shield. The HDM females are larger than the males. Adults can be identified by the genital appearance.

Figure 2 & 3 (p 11-12) are excluded from this web-version due to poor image quality.

Mite developmental stages and population dynamics

Mites have a number of different stages to complete a full developmental cycle (for example from egg to egg). Figure 4 shows the five stages of the HDM life cycle: egg, larva, protonymph, tritonymph and adult (some authors also mention a prelarval stage that takes place within the egg (Hart, 1990)). The reproduction of HDM is exclusively sexual. Adult males mate with adult females (some have also suggested that they mate with female tritonymphs (Hart, 1998)). A few days after mating the females begin to lay eggs at a rate of 1-3 eggs per day (Hart, 1998) however, de Boer et al. (1998) report that *Dp* lay almost 20 eggs per day at 16°C and 76% RH. Each developmental stage grows in size and differs morphologically from the other stages (Hart, 1998). It is thus possible to distinguish between the stages.

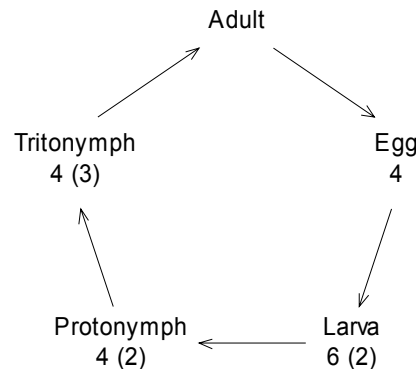


Figure 4. The life cycle of *Dp* at 30°C and 75% RH. The figures show approximate duration in days for each cycle. Numbers in brackets indicate the part of each stage that is quiescent. Note that the data are for optimal climate for HDM development. Adapted from Arlian, (1992).

The adult *Dp* has a life span of 60-80 and 100-150 days for males and females, respectively. However, the females only lay eggs during the first half of their lives (Hart, 1990). The larva, the protonymph and the tritonymph all have quiescent (motionless) stages during which metamorphosis takes place (the next stage develops). The egg can also be seen as a quiescent stage. During the non-quiescent stages the mites are active and feed.

Many invertebrates have dormant stages in which they minimize water loss (Danks, 2000). There are different mechanisms for this, for example high initial water content, vapor tight structures or coatings, and anhydrobiosis (metabolism ceases after a period of preparation). For quiescent HDM a water tight cuticle and lowered metabolism (not anhydrobiosis) seems to be the methods used for survival. The protonymph quiescent stage has also been found to be especially desiccation tolerant (Arlian, 1992). It has a low metabolic rate and can survive for long periods at low humidity. The oxygen consumption of a quiescent protonymph is almost 30 times lower (Arlian, 1992) and they lose water at less than 1% the rate of an active protonymph (Arlian and Wharton, 1974). According to de Boer and Kuller (1997), the quiescent stage of *Dp* is brief and never prolonged as it is for *Df*. This may be one reason why *Df* populations usually are found to be more desiccation tolerant.

It should be noted that the quiescent stages are metabolically active and that, e.g., a quiescent protonymph consumes stored energy reserves (Danks, 2000). It also loses water by evaporation in most environments, although at low rates (it is not believed that mites metabolize, e.g., stored fat, to gain water as some other arthropods do (Danks, 2000)). In a constantly dry environment a quiescent (dormant) stage will die either from desiccation or from exhausted energy reserves.

It is likely that protonymphs become quiescent when the RH drops below the CEH and that these quiescent protonymphs survive long dry periods and serve as the source of breeding mites when the RH is more favorable. As quiescent protonymphs can be seen firmly glued to the substrate (Arlan, 1992), they are probably not easily removed, e.g., by vacuuming.

Dp completes its life cycle (egg to adult) in 123, 34, 19 and 15 days at 16, 23, 30 and 35°C, respectively. *Df* does not develop well at 16 and 35°C (Arlan et al., 2002). Note that because of the complex life cycle of the HDM it is not trivial to design population growth experiments. It is common to start with only female mites (Arlan et al., 1999b) or a mixed population. To see true population dynamics one must probably run experiments for a rather long time, at least a few generations. Many reported measurements have been shorter than this and aimed at measuring, e.g., the survival of female HDM or time to complete an egg to adult “cycle” (but, not including the full adult to egg part that can take up to 75 days during which the females lay eggs).

According to Hay et al. (1992), competition and predation are not important factors in the population dynamics of HDM. The most important predatory mite (*Cheyletus*) found in house dust has an optimum RH at 90%, higher than HDM. If this is true, then HDM successfully inhabit a niche that no predators have succeeded in exploiting. They, who previously lived – together with predators – outdoors in warm, humid climates, have found that in our colder climate they can survive in our buildings, something that their predators cannot do.

Laboratory HDM techniques

There are several rather simple methods for mass culture of HDM, see for example (Miyamoto et al., 1975; Ree and Lee, 1997). These methods include keeping the mites at conditions for optimal growth (about 75% RH, 26°C) and feeding them with nutritious food. The animals must of course be kept in cages with aeration holes of less than a few tenths of a mm to prevent the escape of the mites (or by putting some sticky compound on the rim of the dishes in which they are grown (Hart, 1990)).

Most laboratory studies of HDM population dynamics are made on laboratory cultures, i.e. mites that have been cultured for many generations in the laboratory. The mites in such cultures may show different responses to environmental parameters than do free mites in buildings. Reasons for this may be (Hay et al., 1992) that laboratory HDM are adjusted to optimal RH, optimal temperature, and that they are given more nutritious food than found in, e.g., beds. It has also been seen that laboratory mite cultures are dominated by single species of fungi (Hay et al., 1992). Colloff (1987) found that a 17 year old laboratory culture of *Dp* had significantly longer egg development time and higher egg mortality than did eggs from a wild population.

To start a HDM culture, one should place at least 40 HDM of the same species, but of different developmental stages, in an environment of 75% RH and 25°C (Hart, 1990). Commonly, a Petri dish with a thin layer of suitable food is used (the mites stay at the bottom of the dish unless they are crowded). The cultures can be placed in the dark, or with a daily light cycle. As a general food for all types of mites found in buildings one can use a 1:1:1 mixture of fishmeal, insect meal and dried yeast powder. For HDM one can also use a 1:1 mixture of acetone washed human skin scales and dried yeast powder. Instead of skin scales one can use beard shavings. HDM have also been reared on 1:1:1 of wheat germ, dried liver and dried yeast powder, and 1:1 (vol:vol) of dried *Daphnia* and ground, dry yeast (de Boer et al., 1998) (these diets are free of human products). Brody and Wharton (1970) simply raised *Df* on dog food as we have done. For details on HDM rearing, consult Hart (1990).

For identification and quantification of HDM one usually uses flotation techniques to collect the mites (dead) for direct inspection in a dissecting microscope or for permanent mounting on microscope slides. Techniques for this are described in some detail by Hart (1990). The most common technique for identifying mites is to use a phase contrast microscope (Hart, 1990).

The nitrogen excretion products of HDM and other mites consist of guanine. This can be used to quantify HDM levels.

For studies of mass (water) gain and loss of individual mites (with a mass of 5-10 μg) balances 0.1 μg balances are used, and each measurement is repeated several times. CO_2 anesthesia is sometimes used to stop the animals from escaping (de Boer et al., 1998). Exposures to different RH are made with saturated salt solutions or glycerol-water mixtures in thermostated rooms or cabinets.

Mite allergens

It was Dekker who first suggested that mites were responsible for “house dust allergy”, but it was not until 1964 that this hypothesis was first verified by Voorhorst and co-workers (Fain et al., 1990). Like most common allergens (cat, dog, latex, pollen etc.) the allergenic substances from HDM are natural proteins. The HDM allergens are mainly found on the faecal pellets, but also in and on the animals themselves.

Allergens are identified by the first three letters of the genus and the first letter of the species from which it originates. HDM allergens are thus termed *Der f* and *Der p*. To this one adds roman numerals if there are several allergens from the same source. For HDM allergens, *Der f* I and *Der p* I are considered to be the major allergens, but there are also others, like *Der f* II and *Der p* II (Guérin, 1990). The HDM allergens are related to allergens from other mites and other related species.

The allergens are quite stable to dry heat (Cain et al., 1998). For example does 80°C for 60 min gives no reduction in allergy concentrations, but at higher temperatures a breakdown is seen; 15 min at 120°C gives about a 50% reduction. The *Der* allergens are more sensitive to dry heat than cat and dog allergens. Autoclaving (wet heat) gives a higher reduction in allergen levels, and autoclaving has been suggested as a method to reduce allergen levels. Under natural conditions, mite allergens can be extremely stable (de Boer et al., 1995).

Temperature sensitivity of HDM

The highest tolerable temperature for 24 h of *Dp* is 45.5°C (Hart, 1990). *Df* survival at high temperatures is independent of RH and is about 200, 30, 8 and 4 minutes at 45, 50, 60 and 70°C, respectively (Chang et al., 1998). Mites are killed in laundry washing temperatures above 55°C (de Boer, 1998) or when mattresses are heated to 45°C (de Boer, 1990). It has also been reported that sunlight can destroy a mite population, for example in rugs laid out in the sun (de Boer, 1998), but this is then probably an effect of the UV radiation, maybe in combination with high temperatures.

HDM are also killed by low temperatures. Liquid nitrogen has been used as a way of generating low temperatures in buildings, but mites are also killed in a house-hold freezer (de Boer, 1998). The lower thermal death points for *Df* and *Dp* have been reported as -18°C for 24 h for *Df* and -28°C for 6 h for both *Df* and *Dp*.

Acaricides

There are a number of different chemical treatments available to destroy mite populations. The most common is benzyl benzoate, that has been observed to kill all mobile mites, but the large scale application of benzyl benzoate in buildings have given variable results, probably because it is less effective on the non-mobile stages (de Boer, 1998). Guérin (1990) gives an overview of acaricides.

Dispersal of HDM

HDM spread from building to building by infested clothing and scalp hair (de Boer, 1998). Once in a building, its spread is determined by the indoor climate (or maybe one should say indoor microclimates). Very large differences can be seen between HDM counts in seemingly similar buildings. This is probably the effect of different indoor climates as it is believed that all buildings are continuously contaminated by HDM carried into the buildings.

HDM also have a possibility to spread by walking. Although this is probably not a mode of spreading between buildings, it is a method by which the mites seek the most favorable environments within, e.g., a mattress (de Boer, 1990). Mite populations can move (at least short distances) to areas of higher humidity or higher food supply (Siebers et al., 2004). In a Spanish study of how to control storage mites on ham, it was found that it was no use to dry the meat on the outside as the mites then moved inside the meat and totally disintegrated it (García, 2004). This has not been much studied for HDM, but there is presently a group in Cambridge (UK) working with models on HDM movement in response to temperature and RH (Pretlove et al., 2001). This is certainly a factor that has to be considered in modeling HDM survival, e.g., in mattresses. It may, for example, be easier for the mites to move in a mattress made of textile or animal fibers than in a foam mattress (or vice-versa).

Water balance of HDM

Many studies have shown a significant and positive correlation between the air humidity in a home and the concentration of mite allergens in the house dust (de Boer, 1998; Dharmage et al., 1999). Recent studies have also shown that efficient drying of homes can reduce the allergen concentrations (Arlian et al., 2001).

All organisms are one of two types regarding their ability to control their internal humidity level (Nash, 1996):

- Homiohydric organisms have the capacity to maintain water status at fairly constant levels, usually close to that of pure water (0.990-0.996 (Arlian, 1992)). Examples are most flowering plants and conifers, mammals, insects, and spiders. By different means homiohydric organisms can keep their metabolic processes running even when the ambient environment is dry. Homiohydric organisms usually die if they dry out.
- Poikilohydric organisms, in which the water status varies passively with the surrounding environmental conditions. Typical examples are bacteria, moulds, lichens and some ferns. Many poikilohydric organisms can survive drastic changes of internal water activity, but they are only active when they are humidified by rain, dew, high humidities etc., and dry out when the ambient environment is dry.

HDM are homiohydric organisms and keep a high internal water activity of 0.99 (Arlian, 1992) similar to that of, e.g., humans. This is an impressive achievement as HDM can live in quite dry environments and have no possibility of acquiring liquid water by drinking. They

also face a serious problem in their small size. It is easier to have a positive water balance if one has a larger body as the surface/volume-ratio becomes smaller. As an example, consider two organisms with the same spherical shape, the same internal water activity and the same protective skin. If one has a mass of 100 kg (a human) and the other has the mass of 10 μg (a HDM), they will have approx. radii of 0.3 m and 0.6 mm, respectively. Fractional loss of body mass is proportional to surface area divided by volume, i.e. to the inverse of the radius, so the smaller animal will lose a certain fraction of its body mass 500 times faster than the larger animal. The situation is possibly even worse as larger animals can have thicker protecting skins that further reduce the water loss.

Although insects and spiders are believed to be homiohydric, the springtail *Folsomia candida* seems to have the possibility of acclimate to lower humidities by increasing the concentrations of sugars and polyols when conditions are dry (Sjursen et al., 2001). This is similar to the strategy employed by for example moulds: instead of keeping the water activity high, they try to run their biochemical processes at the lower ambient water activity. Such a capacity has not been seen for HDM. As HDM are extremely well adapted to dry environments, it may be improbable that further desiccation tolerance could be acquired, e.g., by mutagenesis (in a similar way as has been shown possible for the much less desiccation tolerant fruit fly *Drosophila melanogaster* (Telonis-Scott and Hoffman, 2003)).

A reduction of indoor RH could be a way to eliminate HDM problems, but it has been difficult to show this in practice. For example did Hyndman et al. (2000) find no correlation between the use of dehumidifiers and HDM counts. A weakness in this study (and in most other similar studies) is that the RH was measured in the room air and not in the most critical position, e.g., in a carpet or in a mattress. However, one recent study by Arlian et al., (2001) of about 75 homes in Dayton OH, USA, seems to have been successful in showing that reduction of RH is a practical means of reducing HDM populations. In this study, homes that kept an RH of less than 51% had steadily decreasing concentrations of both HDM and HDM allergens. In this study RH was measured in the most humid position in the homes: chair, carpet or bedroom floor (not in mattresses).

Mite water balance

Figure 5 shows all avenues of water gain and water loss from a HDM. The most important loss-route is probably evaporation, followed by defecation/excretion and oviposition (for females). The water loss tolerance of HDM is high. *Df* can lose 52% of its body water without dying (Arlian and Wharton, 1974). Their main way of acquiring water is by active uptake from the air (see below).

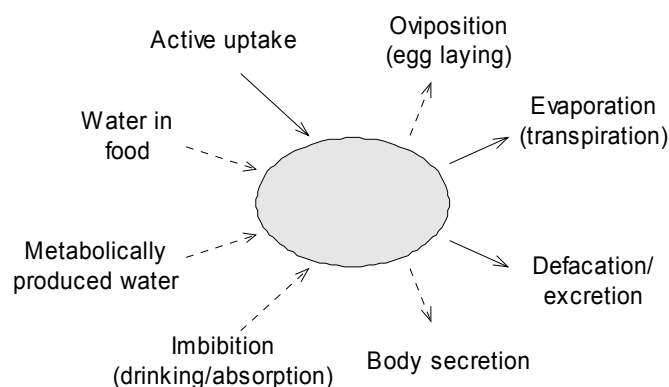


Figure 5. Avenues of water gain and water loss of HDM. The most important routes are shown with solid arrows. Based on Arlian, (1992).

One method of studying body water dynamics in small animals is to give them radioactive (tritiated) water and measure times of uptake or clearance by assessing the radiation from the animals (Arlan and Veselica, 1979). Such studies have, e.g., been made on the booklouse (an insect) by Devine, (1977, 1982), who found that the body water dynamics of a booklouse can be described as that of a single compartment from which water is lost by first order processes and gained by absorption from the air and from metabolism. Such studies on *Df* came to the conclusion that these animals should be modeled by a two compartment models below CEH, but with one compartment above CEH (Arlan and Wharton, 1974). However, after some time of desiccation, it seemed that water was lost from only one compartment, just as above CEH. Probably HDM can be modeled by a one compartment model also below CEH, but with an extra water loss through the glands where the dried hygroscopic fluid does prevent all water losses. An overview of this type of studies of water modeling can be found in (Wharton and Richards, 1978).

Active uptake of water from unsaturated air

It has been known since 1946, that ticks could absorb water vapor from the air, but it was at that time thought that the absorption took place through the cuticle. Rudolph and Knülle (1974), were the first to show that the mechanism of active uptake of water vapor is situated at the mouthparts of ticks (as it is of HDM). A rather large group of arthropods have mechanisms for active uptake of water from unsaturated air. This taxonomically diverse group includes species of isopods, mites, ticks, grasshoppers, cockroaches, lice, fleas, beetles and flies (Danks, 2000). A relatively large number of studies have been made on unattached ticks (Browning, 1953; Knülle, 1966; Lees, 1946; Meyer-König et al., 2001a; Meyer-König et al., 2001b; Strey et al., 1996; Wharton and Richards, 1978) and insects (Devine, 1977; Devine, 1982; Rudolph and Knülle, 1982). One reason why more work has been done on ticks than on mites – apart from that ticks are interesting to medical entomologists as they may spread disease – is that ticks are much larger than mites. Typically, unattached ticks have a mass of 10 mg (Strey et al., 1996), compared to the 5-10 µg mass of a mite. There are much less experimental difficulties involved in experiments on ticks than on mites.

The mechanism of uptake of water from unsaturated air is hygroscopic secretions in oral (ticks, mites, insects) or rectal (insects) sites in some invertebrates. The secretions are salt solutions that are excreted on the outside of the animal where it absorbs water from the air. The solution, now with higher water content, is later brought inside the animal where water is transferred from the solution to the body fluids. This internal drying of the hygroscopic solution is an energy consuming process. Below the CEH the hygroscopic liquid dries out and forms a plug, preventing further flow. In the cases (mainly for ticks) where the composition of this fluid has been studied, it has been found to contain sodium, potassium, chloride and other components (Arlan and Veselica, 1979). Wharton and Furumizo (1977), found that the dried plugs of *Df* contained high concentrations of potassium chloride. As quite different CEH are found for different species it is possible that the hygroscopic solutions used have different chemical composition. Saturated solutions of NaCl and KCl have water activities of 0.75 and 0.85, respectively; quite different from the CEH of less than 60% RH for HDM.

Although the organs responsible for the active uptake of air humidity have been identified in mites, much more detailed descriptions exist for other animals. Rudolph and Knülle (1982), give a detailed account of the uptake mechanisms for a number of insects, for example position and movements of uptake organs during vapor uptake, and CEH (the lowest recorded is 43%). As these insects and ticks are large, e.g., compared to mites, it was possible to block different organs or hinder their movement by applying wax to test which structures that were responsible for the water uptake. Figure 6 shows the result from a measurement on an insect.

Such results gives a clear picture of how such mechanisms may work, but it is not probable that exactly the same mechanisms are active in mites, as mites differ significantly from insects in their morphology.

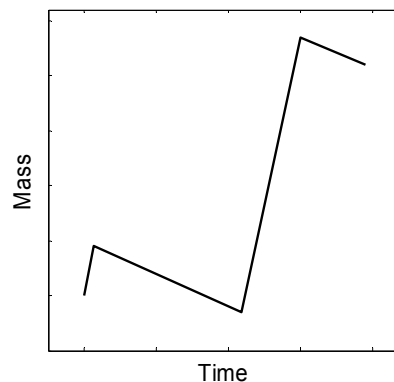


Figure 6. Extract from a weight recording of an insect (*Badonnelia titeri*) in a climate of 93.5% RH and 20°C. Adapted from Rudolph and Knülle (1982) who observed that the mouthparts of the insect were in different positions during weight increase (water uptake) and decrease (water loss by perspiration etc.). A similar result for a feather mite, which has a similar size as a HDM, can be seen in Gaede and Knülle, (1987).

Arlian and Wharton, (1974) found that the active water uptake was at about 0.1 and 0.07 $\mu\text{g}/\text{h}$ for a *Df*, at saturated and CEH-conditions, respectively. Its capacity thus seems to be quite independent of the ambient conditions. Rather detailed descriptions of how this mechanisms may work are found in (Wharton and Furumizo, 1977; Wharton et al., 1979).

Water loss

One of the main water loss paths in HDM is general evaporation of body water that diffuses through the cuticle or in joints between different body parts. It may be difficult to quantify the different routes of water loss from a mite. One possibility is to do as Dautel (1999) did for *Argas reflexus* tick nymphs, and calculate a whole body permeability of 200 $\text{pg}/\text{m}^2/\text{s}/\text{Pa}$. However, it is not probable that this value can be used for the much smaller HDM.

Optimal RH for growth

The conditions most often used in laboratory conditions for optimal growth of HDM is 75% RH at 26°C (Hay et al., 1992). However, *Df* may prefer somewhat drier conditions (Fain, 1990).

Survival at constant relative humidity

The CEH is defined as the lowest RH at which the mites can extract sufficient water from the air to compensate for water lost by transpiration etc. (Hart, 1990). The CEH should be quite close to the RH, at which active uptake up water from the air starts if the uptake takes place at a much higher rate than perspiration (Strey et al., 1996). CEH has been determined to be 58-60% RH, see Table 1 for an overview of results.

Note that even when a HDM is exposed to RH lower than CEH it maintains its metabolism and can be active, although it may have problems with, e.g., eating. There are also reported CEH values of 70-73% RH (Hart, 1990), but these must be wrong, since HDM survive well at lower RH.

Table 1. Measured CEH of HDM.

Der. pteronyssinus		Der. farinae		Reference
CEH / %RH	T / °C	CEH / %RH	T / °C	
58-60	20			(de Boer and Kuller, 1997)
		52	15	(Arlian and Veselica, 1981)
		58	25	(Arlian and Veselica, 1981)
		63	30	(Arlian and Veselica, 1981)
		69	35	(Arlian and Veselica, 1981)

Arlian et al., (1998) found that at 65, 70 and 75% RH *Df* and *Dp* populations doubled every second and fourth week, respectively (there was no clear influence of the RH level). When these thriving cultures were placed at lower RH they declined as seen in Fig. 7. It is seen that the *Df* population initially increased at 45 and 50% RH due to egg hatching. At lower RH a continuous decline was seen. Laboratory studies show that *Dp* has slightly higher RH requirements than *Df*, and that the latter better survives long periods of low relative humidity (Arlian, 1992).

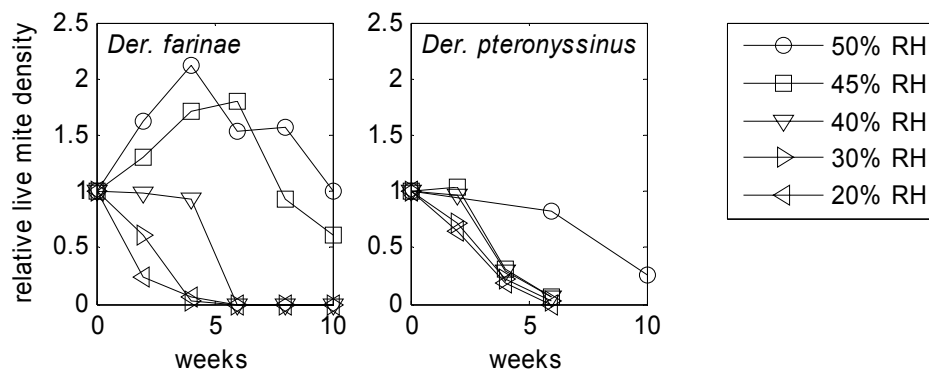


Figure 7. Survival of *Df* and *Dp* at low RH. Data from Arlian et al., (1998).

Colloff, (1987) found that HDM eggs hatched at every combination of RH and temperature between 55-100% RH and 10-35°C, respectively. Optimal conditions were 35°C and 80-85% RH, i.e. much higher than for HDM population growth. It should be noted that HDM cultures do not like too high RH. Arlian et al., (1998, 1999b) found that continuous exposure to 85% RH inhibited population growth at 20-22°C. It is possible that competition from fungi and other organisms increase at higher RH, the decline seen by Arlian et al., (1998) was caused by mould growth.

CEH values as a function of temperature can be compared with actually measured RH(*T*) values. If CEH is less than RH(*T*) a HDM population should not be able to establish itself. One example of such an exercise is given by (de Boer and Kuller, 1997) that found that directly under a sleeping person the absolute humidity increases, but the RH increase is small as the temperature also increases. In their measurement the RH was below the CEH,

essentially all the time. However, there may be other zones in a bed where the conditions are more favorable for HDM growth.

Cunningham, (1996) gives a compilation of doubling/halving times for populations of *Df*, *Df* and *Euroglyphus maynei* at constant conditions based on results from four sources.

Survival at fluctuating relative humidity

As it was noted that, although HDM cannot survive continuous exposure below CEH, they can survive when the *mean* RH is below CEH, it was of interest to make studies at fluctuating RH conditions. A second reason is that RH conditions in beds and carpets, where the HDM thrive, are fluctuating.

de Boer and Kuller, (1997) subjected *Dp* cultures to 10% RH, except for 0, 1.5, 3, 6 or 12 h per day when they were exposed to 90% RH. It was found that all cultures that received 1.5 h or more of daily high humidity had a higher survival. Only 1.5 h per day led to a decline in the number of mites, but 3, 6 and 12 hours allowed reproduction to take place, and 6 and 12 hours gave population growth similar to continuous 75% RH.

de Boer et al., (1998), in a similar study as above, exposed fasting mites at 16°C to 36% RH interrupted by daily periods of 76% RH. With only 1.5 h of 76% RH, more mites survived a 10 week exposure than if the RH was constantly 36%, although there was no reproduction. With 3 or 6 h of 76% RH almost all mites survived.

Arlan et al., (1998) exposed *Df* eggs to daily cycles of 75 and 0% RH and found that larva emerged when the higher RH level was 2 h or longer. To complete a life cycle they needed 6 h of the higher RH level, but almost half the larva or nymphs survived the drier regimes for 70 days.

Arlan et al., (1999a) studied the life cycles of *Df* exposed to cycles of RH levels 35 and 75%. They found that life cycles were completed (egg to adults) when the HDM were given 4 h per day or more of the higher RH (Table 2). They found no significant differences in development times for the different sexes, except for the 6/18 h cycle where the male mites had a total life cycle time of 58 days compared to 70 days for the females. The times for egg development for *Df* in the table can be compared with the lower values 8-10 days reported by Colloff, (1987) for egg development time at 20°C for *Dp* at constant RH in the range 55-100%. Note that the percent of the eggs that completed a life cycle in the experiments were 76, 60, 70, 49 and 0 for the five exposure types given in Table 2.

Table 2. Time to complete a life cycle for *Df* under fluctuating daily RH conditions at 21°C. Mean values for male and female *Df* from (Arlan et al., 1999a). The values in () are the part of each stage that was quiescent. The larva that emerged from the eggs at 24 h 35% RH died in about 5 days.

Time at each RH level / h		Time for each development stage / days				
35% RH	75% RH	Egg	Larva	Protonymph	Tritonymph	Total
0	24	14	12 (4)	8 (4)	8 (4)	41
16	8	15	19 (6)	12 (5)	11 (5)	58
18	6	15	21 (7)	15 (6)	13 (5)	65
20	4	16	30 (8)	20 (7)	16 (6)	82
24	0	14	-	-	-	-

Arlian et al., (1999b) studied population growth of *Df* when RH alternated between a value below (0 or 35% RH) and a value above (75 or 85% RH) CEH. The results are summarized in Fig. 8. At 75% RH the populations increased monotonously, but at 85% RH long term growth was not seen (possibly because of fungal interactions). With 0% RH 22 hours per day and 75% for only two hours, all mites died, but at all other tested combinations of high and low RH, a population survived the experiments. It can be concluded from this, and the other studies mentioned above, that HDM only needs brief spells of high RH to survive in climates below CEH.

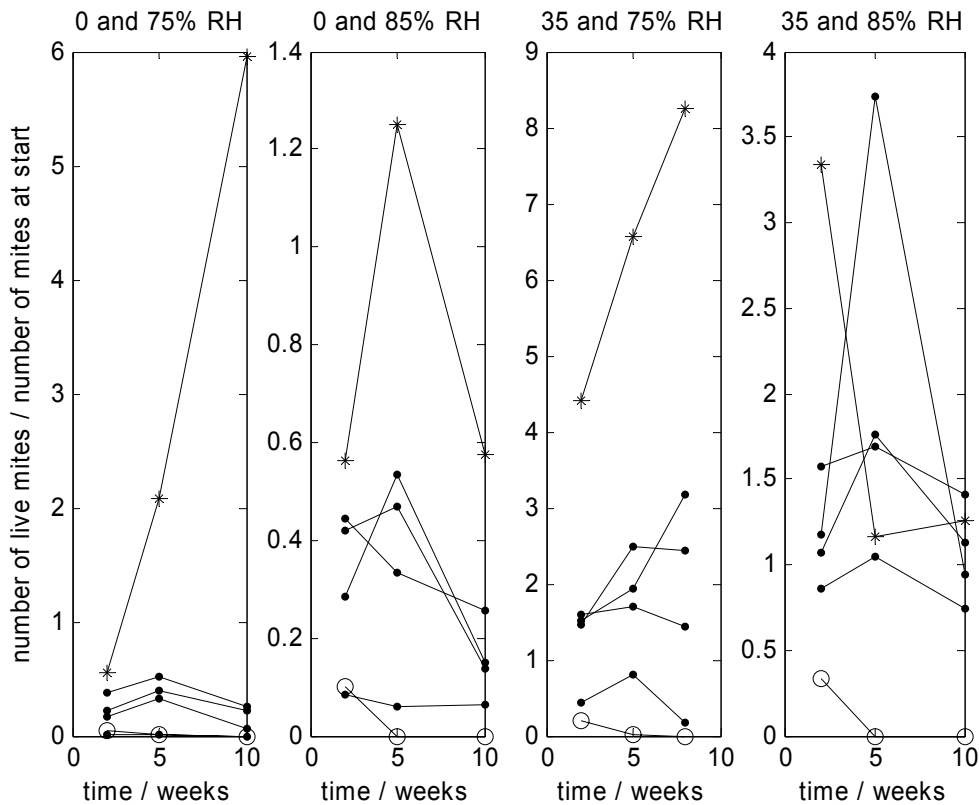


Figure 8. Results of studies of population dynamics at fluctuating RH conditions. Based on experimental data from (Arlian et al., 1999b). Each subplot represents a combination of high and low RH (see top of subplots). Data are given for continuous exposure at the high level (stars), continuous exposure at the low level (circles), and for four fluctuating conditions (solid dots): 2/22, 4/20, 6/18 and 8/16 (hours low RH/hours high RH). Except for a few data points, longer times at high RH gives higher population. See (Arlian et al., 1999b) for details of the experiment, standard deviations etc.

HDM, beds and bedrooms

House dust mites are mostly found in beds, upholstered furniture and carpets and their activity and viability is, as mentioned in the earlier section, dependent on the microclimate of these habitats, especially the temperature and humidity plays an important role. The hygrothermal conditions of these habitats are governed by the moisture balance in the room and the moisture balance of a room is determined by the outdoor climate, the moisture supply, the moisture buffering from surface materials of the room and furnishing, the ventilation rate, the possible condensation at surfaces and the variation of these parameters with time (Svennberg, 2003). The daily variations in temperature and humidity are to a large extent determined by the behavior and habits of the occupants (Rode et al., 2001). The moisture buffering capacity of

materials in the indoor environment will dampen the RH variation of indoor air, at the same time the moisture buffering will give a variation in humidity in the materials, for example in a carpet or a chair (both which are possible house dust mite habitats). Figure 9 describes the hygrothermal flow chart, which can be used to estimate the risk for production of humidity related bio-contaminants such as mite allergens (Künzel and Kiessl, 1997).

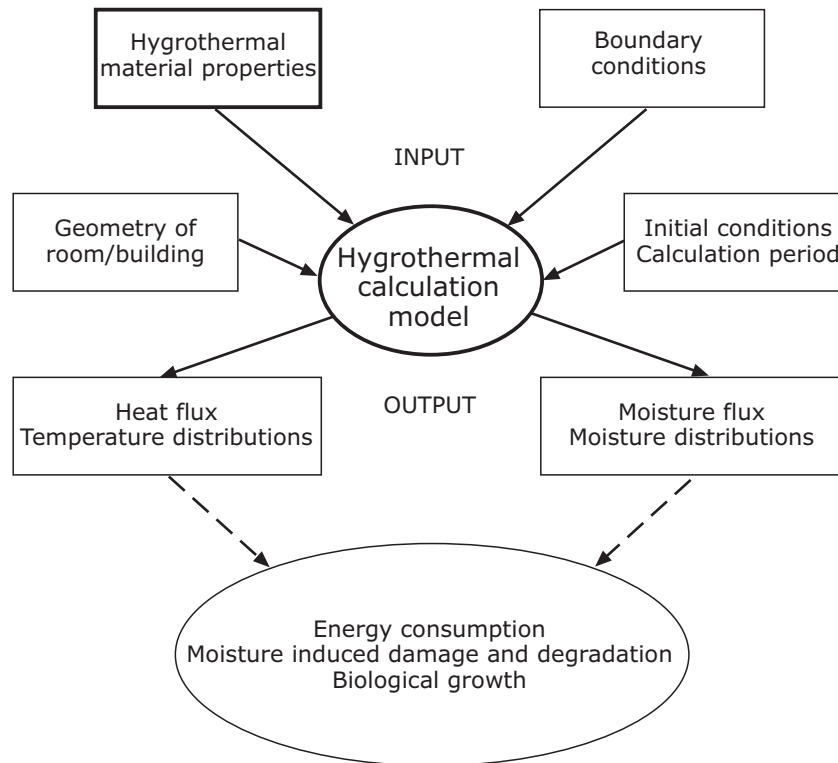


Figure 9. A hygrothermal flow chart, describing the input needed for hygrothermal simulations of a building and the expected use of the output. (Künzel and Kiessl, 1997)

The moisture balance in a Swedish perspective

Since all the factors determining the moisture balance in a room vary with geographical position, building characteristics, occupant behavior and furnishing preferences a wide spread in the hygrothermal conditions in the microclimates relevant for house dust mites are found globally. This project is concerned with mite reduction in bedrooms in the Swedish context and climate. The outdoor climate in Sweden has large variation, both geographically and seasonal (Harderup, 1995; Johansson, 2004). Typically, in southern Sweden (where mite allergen concentrations are highest) the outdoor mean temperatures are in the order of 0°C in the winter and 15°C in the summer. Further north the winter temperatures are lower and the indoor relative humidity consequently also lower.

The moisture supply is assumed to be composed of two parts. A relatively constant basic supply from plants, inhabitants and pets and short moisture supply pulses that change almost momentarily. These short moisture supply pulses come from, e.g., cooking, washing, showers and baths, activities that are carried out during a shorter period of the day. The moisture supply is assumed to change uniformly within the entire room volume regarded. The variations in moisture supply and moisture production in Scandinavian dwellings have been studied by for example Gustavsson, (2004); Norlén and Andersson, (1993). The average moisture supply found by Gustavsson, (2004) was approximately 2,3 g/m³.

The ability of the interior surfaces to buffer moisture variations of the indoor air is described with a moisture buffer capacity for each material or material combination. Both the surfaces of the inside of the building envelope such as ceilings, floors and walls as well as the furniture and other furnishing, will give an impact on the moisture conditions in the room. For example Plathner and Woloszyn, (2002) have shown that the correlation between simulated and measured moisture conditions of the indoor air is much better if the sorption of interior surface materials is taken into account (Fig. 10).

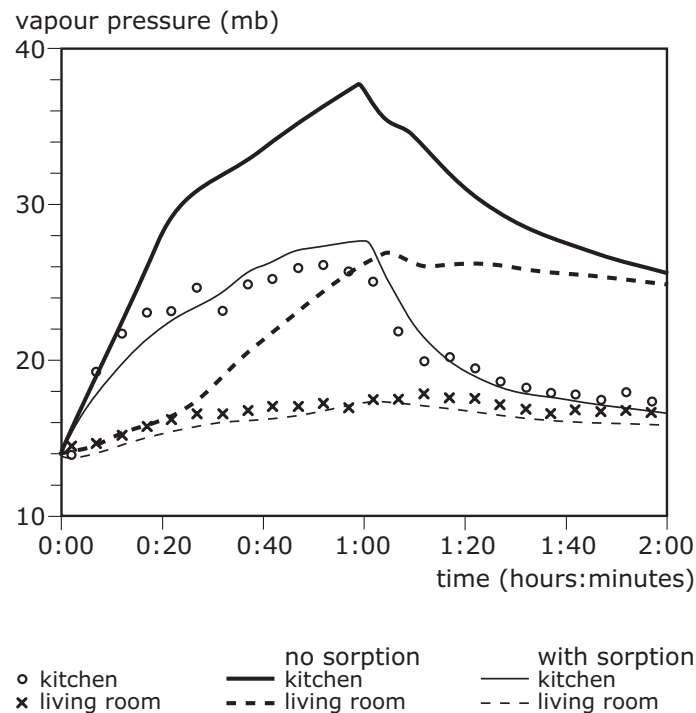


Figure 10. Comparison between measured and calculated moisture conditions of the indoor air. Calculations with the sorption of the surface materials taken into account show better correlation with measured values than the calculations where the sorption was not taken into account. The temperature was set to 24 °C for both calculations and field measurements. Adapted from Plathner & Woloszyn (2000).

The ventilation is crucial for the moisture condition of indoor air. The Swedish Building Code has a minimum requirement of 0.5 air changes per hour (BBR, 2002). Several studies have shown that this level is not reached for the building stock as a whole. The ongoing DBH study in the county of Värmland have made a survey of 390 homes – both apartments and detached houses - and found an average air change rate of 0.36 l/h and 0.47 l/h respectively (Gustavsson, 2004). The impact of the ventilation on the indoor vapor content depends on the air exchange rate, the vapor content of the outdoor air, the moisture supply rate and moisture buffering capacity of the materials present. A high air exchange rate will control the RH of indoor air to a higher degree. The distribution of different ventilations systems from 390 houses investigated in the DBH study is shown in Fig.11.

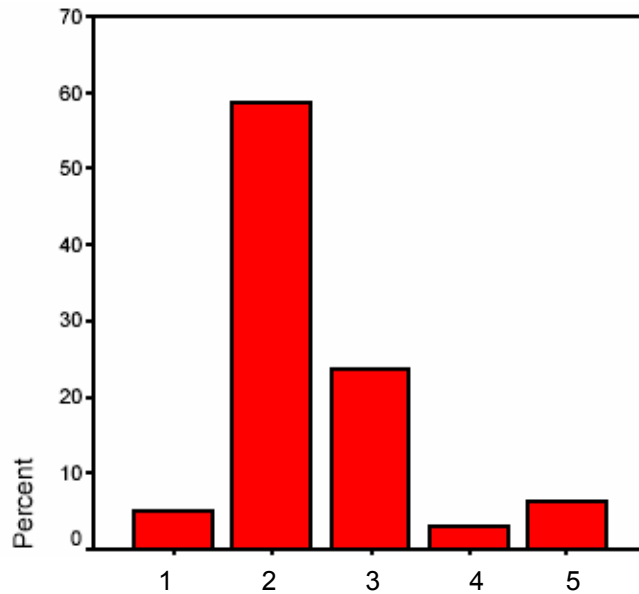


Figure 11. Types of ventilation systems. 1. Natural ventilation without kitchen fan; 2. Natural ventilation and kitchen fan; 3. Exhaust ventilation; 4. Balanced ventilation; 5. Balanced with heat exchanger (Gustavsson 2004).

In Sweden central heating is predominant, and the relations between different heating sources have been investigated by for example Gustavsson, (2004), see Fig. 12. The mean indoor temperature was found to be 20.9 °C.

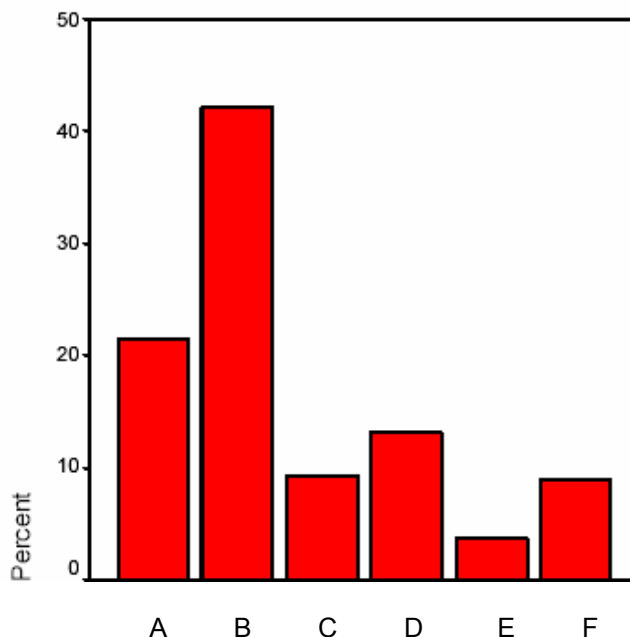


Figure 12. The main heat sources in the homes (Gustavsson, 2004). A. Oil; B. Electrical heating; C. District heating; D. Wood; E. Wood pellets; F. Other.

The age of the home does not necessarily affect the moisture balance, but different technical solutions have different spread within the building stock, often depending on the age of the house. The age distribution of the homes in the DBH study is seen in Fig. 13 (Gustavsson, 2004).

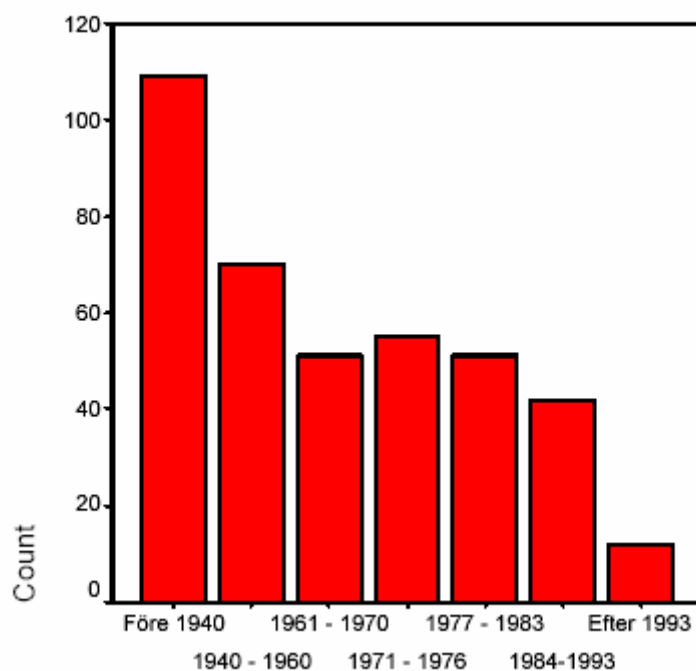


Figure 13. Construction dates of the 390 houses in the DBH-study in Värmland, Sweden (Gustavsson, 2004).

Field measurements

Building characteristics and correlations with mite allergens / mite density

All over the world there have been studies to correlate different building characteristics with the level of mite allergens and/or the density of mites. Since building traditions, outdoor climate and furnishing preferences vary around the world, it is hard to generalize the findings in these investigations, and since the investigations seldom are made double-blind there might be confounders due to the occupants behavior, such as an increased cleaning (Platts-Mills, 2004). Another problem with most of these studies is that the indoor climate have most commonly been measured in the center of the room; the habitats where the mites live (beds, carpets, furniture) probably have quite different microclimates (Cunningham, 1998; Sheikh et al., 2001). Yet another reason for a large spread in results concerning building characteristics and correlations with levels of mite allergens and mite abundance, is the fact that the most studies use questionnaires to assess the building characteristics, giving a large uncertainty in the description of the building characteristics. Only a few studies like Gustavsson, (2004, an ongoing study), Garrett et al., (1998) and Couper et al., (1998) have used home surveys to assess the building characteristics. There are also various definitions of dampness in use (Bornehag et al., 2001), making that characteristic very hard to evaluate in inter-study comparisons.

A nationwide survey in 831 US beds was reported by Arbes et al., (2003). Dust mite allergens were determined from dust samples from the bed. Significant building characteristics for prediction of high levels of mite allergens were the age of the home, house type, mildew odor and higher RH (especially in the bedroom). Other studies have found similar building characteristics to be important (Chan-Yeung et al., 1995; Chew et al., 1999; Couper et al.,

1998; de Andrade et al., 1995; Dharmage, 1999; Emenius et al., 2000; Garrett et al., 1998; HA Mamoon RL Henry JE Stuart, 2002; Harving et al., 1994; Korsgaard, 1983; Martin et al., 1997; Schei et al., 2002; Warner et al., 1999; Zock et al., 2002). However, there is no consensus on if many of these building characteristics successfully can predict higher occurrence of mites and mite allergy symptoms. High RH being one exception and low air exchange another. It shall be noted that not all studies have made air exchange measurements and that the spread in sampling method and sampling period for RH and temperature vary to a high extent.

The mite density in dust-sensitive patients homes was monitored for 2 years (sampling with 3 week intervals) by Arlian et al., (1982) and correlated to indoor climate and physical factors of the indoor environment. The significantly highest mite levels were found in the most used upholstered furniture with fabric, and in the carpet of the living room and bedroom. Floors with carpet had significantly higher mite levels than non-carpet floors. The study did not find mattresses to be the main habitat for the mites. The cleaning frequency or thoroughness did not affect the mite levels, and the mite levels were not significantly reduced by successive vacuum cleaning. A seasonal variation in mite density was also seen, with a peak in the humid summer months.

The methods used to sample dust vary. Different levels of mites and mite allergens will be found if airborne dust, settling dust or reservoir dust is sampled (Tovey et al., 2003). This also makes inter-study comparison more difficult.

Mattress type and correlations with mite allergens / mite density

There is no conclusive result on which mattress type that gives the lowest mite allergen levels.

Wickens et al., (1997) have found significantly higher mite allergen levels in kapok and inner spring mattresses and when wool underlay have been used. Garrett et al., (1998) have found similar results. Schei et al., (2002) on the other hand, found the risk to find mite faeces (major source for allergen) to be 4 times higher in foam mattress than in spring mattress.

Pillows and correlations with mite allergens / mite density

Siebers, (2004) has made a study on mite permeability of pillow covering, showing that a more open covering let more mite and dust into the pillow, creating more favorable living conditions. The findings on mite allergen levels in feather pillows and synthetic pillows are not conclusive (Chan-Yeung et al., 1995).

Material properties

There is a great lack of material properties for beds, beddings and other types of textile furnishings (Pretlove et al., 2001; Svennberg, 2003). Material properties that could be useful in investigating the hygrothermal performance in relation to mite occurrence are: water vapor sorption isotherms, water vapor diffusion coefficients, thermal conductivity, and heat capacity. It would also be interesting to devise a measure of the ease at which the mites can move inside or through materials.

Microclimate

It seems to be the microclimate that to a high extent governs the life and activity of house dust mites. It is therefore essential to assess the microclimate in possible mite habitats (Colloff, 1998; Cunningham, 1998; Cunningham, 1999; Sheikh et al., 2001; Strachan and Sanders, 1989). To get a good resolution of the microclimate measurements, small RH and temperature sensors need to be used (Baker et al., 2004). There have been some studies using various

types of small RH sensors, giving a better insight in the climatic differences between the mite habitats and the entire room (Baker et al., 2004; Cunningham, 1998; Hokoi, 2004; Pretlove et al., 2001; Svennberg et al., 2004).

The micro climate in beds have been studied and modeled by several researchers (Cunningham, 1999; Cunningham et al., 2004; Hokoi, 2004; Pretlove et al., 2001), and good agreement between the measurements and the models have been found. It have also been shown that during night time, the hygrothermal condition underneath the sleeping person is not favorable to the mites. Even if a sleeping person releases water vapor the combined effect of this and the high temperatures is a lowering of the RH.

Measurements of the hygrothermal conditions of upholstered furniture have also been undertaken by Baker et al., (2004), Hänel et al., (1997) and Svennberg et al., (2004). Hänel et al., (1997) showed that the upholstery transport moisture mostly in the surface layer below a certain point of compression. It might be, that the compression of upholstery and mattresses (Sukigara et al., 1996) need to be taken into account for a correct assessment of the moisture transport.

Carpets, especially wall-to-wall carpets, have found to be a major mite habitat. Many researchers (Baker et al., 2004; Cunningham, 1998; Cunningham, 1999; Cunningham et al., 2004; de Boer, 2003) have made measurements of the hygrothermal conditions in various types of carpets. Baker et al., (2004) and Cunningham, (1999; Cunningham et al., 2004) have also modeled this micro climate with a good agreement with measured data. Note that wall-to-wall carpets are very unusual in Sweden. Swedish homes have hard floors (PVC, linoleum, parquet, solid wood, tiles), often partly covered with smaller loose?? carpets.

The effect of airing through window opening can be calculated by a model proposed by Nordquist, (2002).

Population / Activity model

Using the modeled or measured micro climate of the mite habitats, attempts have been made by Baker et al., (2004); Pretlove et al., (2001); and Wilkinson et al., (2002) to model the population growth or the activity of the mites. However, more field measurements and laboratory work remain, before these models can reliably predict the mite density or the mite allergen levels.

Humans

The hygrothermal behavior of humans in sleep can be calculated using Fangers Thermal comfort equation (Holmér, 2004) as standardized in ISO7730. The movement during sleep influences the temperature and moisture transfer from the human body to the bed (Hokoi, 2004). The significance of these variations for the moisture condition of the bed and how it will affect the mites need to be investigated more carefully. The clothing of persons may play an important role in the mite infestation of a new house. The clothes are also a habitat for mites (De Lucca et al., 2000).

Reduction strategies

Reducing mites from the indoor environment is a hard task, and requires more than a once-and-for-all measure to be successful (Boner et al., 2002; Fernández-Caldas, 2002; Platts-Mills, 2004). Physical measures seem to be more successful than chemical.

Humidity reduction control strategies

Ten dwellings with mechanical ventilation with heat-exchange in Wellington, New Zealand, were studied by Crane et al., (1998). The result showed that even if the RH was lowered considerably by these mechanical ventilation units, the levels of mite and mite allergens remained unaffected. A closer analysis of the RH levels showed that the RH was lower than CEH only in 39% of the total 24-hour periods under which the measurements were performed.

Arlian, (2001) made an intervention study with 3 groups with approximately 20 houses in each group. Group 1 maintained low humidity (< 51 % RH) with a high-efficiency dehumidifier and air conditioning. Group 2 used air conditioning (and in a few cases also dehumidifier) but did not maintain an RH level under 51 % RH. Group 3 controlled the indoor climate through window opening and airing. This group had an RH greater than 51% RH. In group 1 the allergen levels dropped during the study, the other two groups had seasonal peaks. After 17 months the homes in group 1 exhibited 10 times lower allergen levels than the humid homes of group 2 and 3.

Mechanical ventilation and dehumidifiers

The findings from several investigations employing mechanical ventilation and/or dehumidifiers (Hyndman et al., 1994) as means to lower the mean RH of the house or the room, and thereby reduce the mite density or the mite allergen levels, are inconclusive. Fletcher et al., (1996); Hyndman, (2000); Singh et al., (2002) find no or very small positive effects of the measures taken. Arlian, (2001); Emenius et al., (1998); Harving et al., (1994); Harving et al., (1994) all see a clear positive effect of the mechanical ventilation or dehumidification.

Laundry

Laundry as a mean of mite reduction from clothes and bedding was experimentally investigated by Arlian et al., (2003). It was shown that laundry both reduced living mites on mite-infested textiles and that mite-free items could be infested by the mite-infested textiles in the laundry process. The best reduction was found for detergents in combination with sodium hypochlorite bleach. Similar results have also been found by Causer et al., (2003); Tovey et al., (2001). The positive reduction effect of laundry and mattress cleaning has also been shown in field measurements by Rao et al., (1975).

Heat / Steam treatment

Htut et al., (2001) made a double-blind study on eradication of house dust mites using hot air and hot steam. Two out of three groups had their mattresses and beddings treated with this method. The third group's mattress and bedding were sham treated. One of the treated groups also had a mechanical ventilation unit installed in the bedroom. The results show that this one-time treatment had good effect on eradication of the mites, but only the group with additional mechanical ventilation had low mite levels long term.

Mattress encasings

A common recommendation (The Swedish Asthma and Allergy Organisation, 2004) for house dust mite avoidance is the use of bed encasements. There have been a number of studies the last years, most of them showing a significant but modest effect of the use of encasements (Holm et al., 2001; Kniest et al., 1998; Mahakittikun et al., 2003; Mahakittikun, 2003; Mahrshahi et al., 2003; Peroni et al., 2004; Recer, 2004; Tempels-Pavlica et al., 2004; Tobias et al., 2004; van Strien et al., 2003). There are a number of different materials used in bed encasements and they protect against mites and mite allergens in different ways. The various fabric also have different air and vapor permeability (Kniest et al., 1998). The significance of the moisture properties of the complete bed need to be more carefully investigated.

Dust avoidance

A study by Walshaw and Evans, (1986) shows that it is possible to succeed with dust avoidance procedures in the homes of already allergic persons. The dust avoidance procedures were supplemented by the use of bed and pillow encasings.

Mosbech et al., (1988) used an electrical blanket during day time in a study to reduce mite allergen levels. It was found to be useful a measure to reduce mites in beds.

3. Results from experimental studies of house dust mites

We have tested two methods to study the influence of temperature and relative humidity on HDM. Firstly, we used isothermal calorimetry (measurements of heat production rate) to measure the activity of HDM placed in different climates. Secondly, we counted HDM both visually and by image analysis.

HDM used in the present study

The *Dermatophagoides farinae* (HDM) used in the present study was a generous gift from Allergon AB (Ängelholm, Sweden). They were acquired as a pure culture and were mixed by us with dog food.

Different kinds of experimental techniques to study HDM needs different amounts of mites. Typically a microscopy study needs less than 100 animals, whereas the present calorimetric experiments used 100 mg HDM. As each HDM has a size of about 500 µm and a mass of 5-10 µg (Arlian and Wharton, 1974) each ampoule contained 10000-20000 HDM at start. However, the calorimeters used were not of highest sensitivity. The following is an estimation of how many HDM that would be needed for measurements in different types of isothermal calorimeters:

*Thermometric TAM Air, TAS** 20000

Microcalorimeter (Thermometric TAM) 1000

Nanocalorimeter 20

*used in present study

The counting of HDM, e.g., in Petri dishes as was done in the present study, makes use of much less number of animals, and can also give information about their developmental stages. However, manual counting of HDM is laborious and is not always an exact science as animals can hide, for example under food particles. The image analysis method described below is an interesting alternative to manual counting.

Calorimetric method of measuring HDM activity

Background

Calorimetry is the measurement of heat and heat production rate (thermal power). This is a general method to measure rates of many different types of physical, chemical and biological processes as nearly all processes gives of heat. We use isothermal calorimeters in which the sample on which the thermal power is measured is kept at constant temperature.

In biology isothermal calorimetry has been used on many different types of samples, e.g. (taken from Li (2004)):

- Applied studies in biochemical engineering (Duboc et al., 1999)
- Fundamental studies of yeast metabolism (Larsson and Gustafsson, 1999)
- Aquatic animals, such as fish and insects (Lamprecht and Schmolz, 1999)
- Animal physiology and bioenergetics (Hand, 1999)
- Animal tissues (Kemp and Guan, 1999)
- Human cells (Monti, 1999)
- Plant metabolism (Criddle and Hansen, 1999; Criddle et al., 1991; Criddle et al., 1988; Hansen et al., 1997; Hansen et al., 1995)
- Vegetable respiration (Gomez et al., 2004; Wadsö et al., 2004b)
- Microbial degradation of food stuffs (Shiraldi et al., 1999)
- Mould and rot fungi (Wadsö, 1996; Wadsö, 1997; Wadsö et al., 2004a; Xie et al., 1997)

In most of these examples the biological samples are under aerobic conditions, i.e. consuming oxygen and nutrients, and producing carbon dioxide, water and possibly biomass. Such aerobic processes have been shown to produce about 469 kJ of heat per mol O₂ consumed, and thermal power can thus be used as a measure of respiration rate. We have used isothermal calorimetry in this study as a convenient way of assessing overall HDM activity (respiration).

Calorimetric ampoule

For the whole experimental period the HDM were kept in sealed 3 ml calorimetric glass ampoules with an outer diameter of about 14 mm (shown in Fig. 14). The sample consisted of 100 mg HDM and 500 mg dog food of particle size <1 mm. To prevent the HDM from escaping the top of the ampoules were sealed with a thin double tape (shown black in Fig. 14) and a circular disk of a Millipore filter (hole size 0.22 µm). Water vapor, carbon dioxide and oxygen could thus pass into or out of the ampoule while the HDM stayed inside.

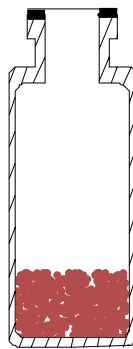


Figure 14. The 3 ml calorimetric glass ampoule used in the calorimetric experiments.

Before each measurement in the calorimeter the ampoules were sealed with an aluminum crimp cap and a Teflon coated rubber seal (standard method in analytical chemistry). This prevented any gas or vapor exchange during the measurements (esp. water vapor vaporization causes large thermal powers and must be prevented). This seal was removed after the calorimetric measurements. When the sample ampoules were in the climate boxes they were only sealed with the permeable Millipore filter.

In ampoules with living and active HDM some were always seen moving around on the glass surface above the dog food. On all tape surfaces exposed to the inside of the ampoule HDM got stuck. When the caps were removed the tape and Millipore was sometimes separated slightly so that some HDM could move into the gap and get stuck there. In this way the HDM could form a bridge over the seal, and eventually escape from the ampoules. When this happened the seals were replaced with new seals and the outside of the ampoules (and the boxes) were cleaned of all free HDM.

During the present exposures in climate boxes, three samples with HDM and one with only dog food was used for each of the ten climates. The samples in calorimetric glass ampoules were exposed according to the following scheme:

- 25 days at 20°C and 75% RH (optimal conditions), first in storage beakers, then in calorimetric ampoules
- 36 days in climate boxes at different relative humidities and temperature programs (with logging of T and RH as described above). During the end of this period the salts in three boxes dried out.
- 15 days at the same climates (no logging)

Calorimetric measurements

All measurements reported here were made in a prototype of a newly developed eight channel isothermal heat conduction calorimeter called TAS. As this instrument has been developed under a secrecy agreement (for use in a field quite different from biological calorimetry) no performance data can be given here. However, the instrument is quite similar to the 20 ml TAM Air (Thermometric AB, Järfälla, Sweden), except that it has 3 ml ampoules.

Figure 15 shows the result of a measurement (after correction for baseline and calibration coefficient) made during the first phase of the measurements when the HDM were all kept at optimal conditions (75% RH and 20°C). It is clearly seen that there is a measurable difference between the seven sample with the same mass of HDM and the blank sample with only dog food. As the measurements continued the difference between samples with and without HDM became smaller and the differences between them were less obvious.

All calorimetric data on HDM activity was taken 120 minutes after the samples were charged into the calorimeters. It is seen in Fig. 15 that the thermal powers are not perfectly constant, so the rather arbitrary 120 minutes does slightly influence the generated results. However, it is normal that calorimetric data of this type shows slight decreases during measurements. One possible explanation for this is that the oxygen levels decreases in the ampoules (see Appendix A).

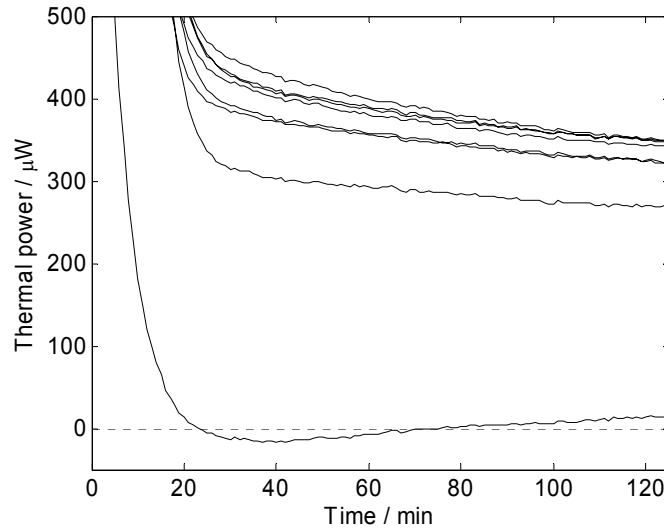


Figure 15. Example of calorimetric measurement on eight samples, one without HDM (only containing dog food).

During these measurements it was seen that both calibration coefficients and baselines of the new TAS instrument are more irregular than TAM Air baselines. The reason for this is not known yet. One set of calibration data (calibration coefficients and baselines) made before the measurement series differed to one taken after the measurements. The differences were up to 5% in the calibration coefficient and about 20 μW in the baselines. Especially the latter is a high uncertainty when the sample thermal powers are in the range 0-400 μW . To choose the best calibration data set the following procedure was used. It was assumed that the samples with only dog food should give close to zero thermal power. Evaluations of all data were therefore made with the two calibration data sets and it was found that the result (values close to zero for samples without HDM) was much better when the calibrations data set taken after the measurements were used. These have therefore been used in the present evaluation. This was a reasonable procedure because – as the samples were randomly assigned to calorimeter channels – any random error should make the result less good. The best result (giving the result expected, giving lower random noise etc.) can be expected to be generated by the most true set of calibration data. In the future more calibrations should be made so that one is able to remove erroneous calibration data.

The calorimetric measurements is a prototype for large scale measurements with isothermal calorimetry. The main idea is to make many measurements with little work. Figure 16 shows the most important parts of this type of set-up that was used for the second time at Building

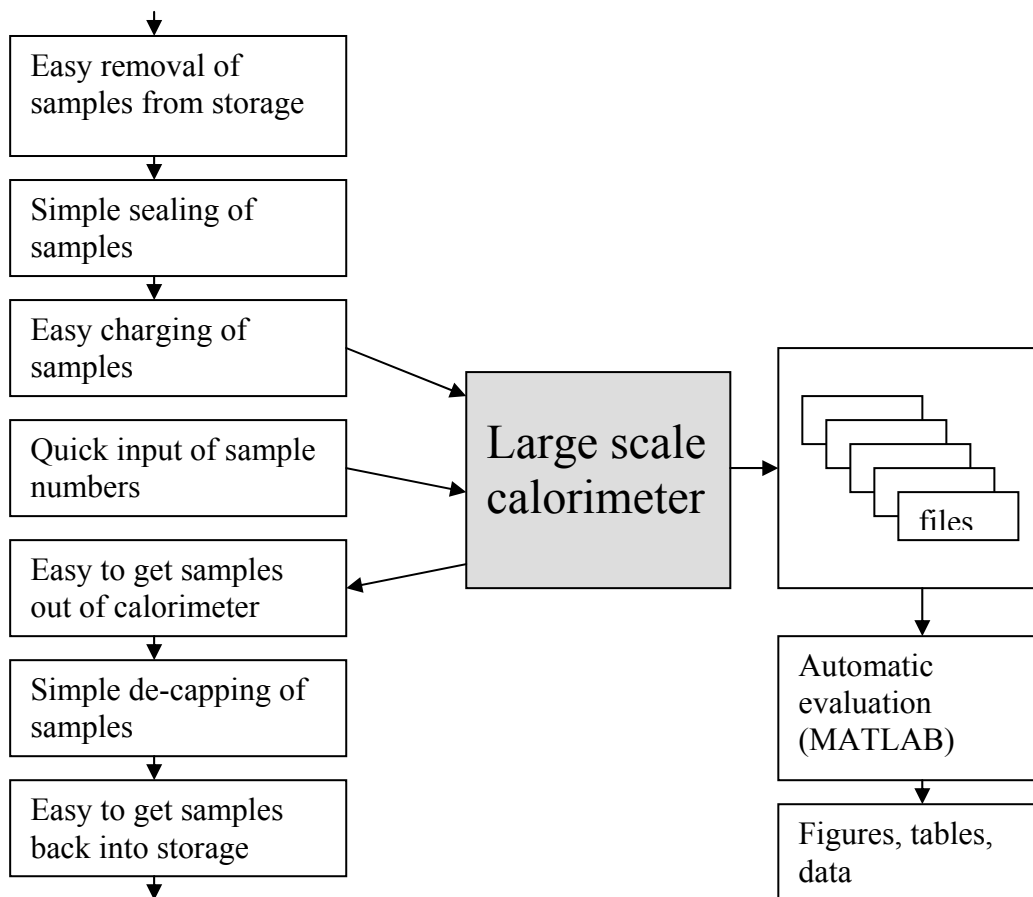


Figure 16. The idea of Large Scale Calorimetry as a method to quickly generate large amounts of data.

Materials calorimetry laboratory (first time was in a study on a correlation between integrated calorimetry measurements and ergosterol concentration reported by Li (2004)). Although there have been some problems in the present study, the idea works well, for example:

- Removing samples from storage, capping and charging them and entering their sample numbers took about 10 minutes for eight samples. With improved capping this figure should decrease to 10 minutes for 32 samples.
- The eight channel TAM Air works well, the used eight channel TAS needs some further development to increase its sensitivity.
- Automatic evaluation with MATLAB programs has worked well and is very flexible.

Results of calorimetric measurements of high temperature exposure

As a test of the calorimetric method we made an experiment with eight ampoules with HDM previously measured in the calorimeter, in which we exposed the ampoules at 45°C in a heating block for time periods from 0 to 180 min. One day after this exposure the ampoules were once more measured in the calorimeter to assess whether the high temperature exposure had damaged the HDM. The result is shown in Fig. 17. It is seen that there is a good correlation between exposure time and survival ratio (defined as ratio of thermal power after and thermal power before exposure). Measurements were also to be done with low

temperature and oxygen depletion, but the thermal powers of the samples decreased to such low values before it was possible to perform these measurements.

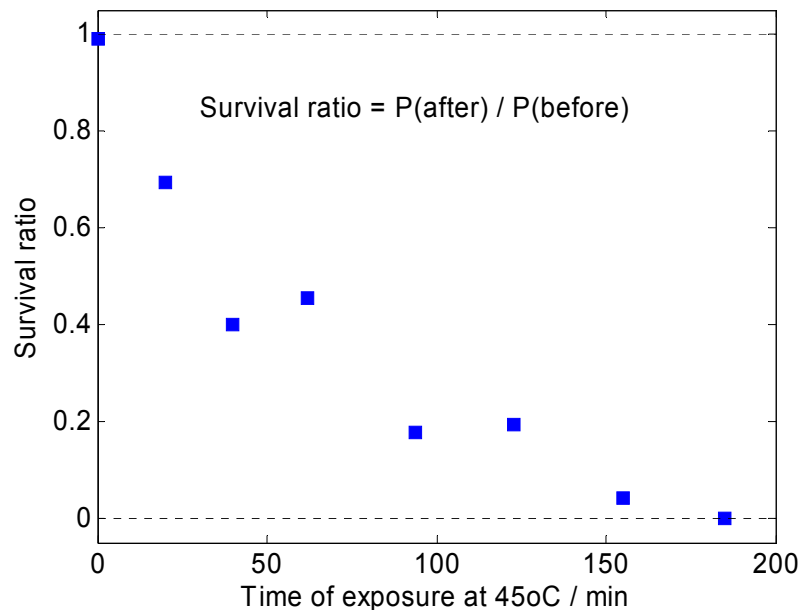


Figure 17. Results from measurements with exposure of HDM at 45°C.

Results of calorimetric measurements as a function of climate

All results are shown in Figs. 18 and 19. It is seen that all samples has thermal powers of 200-500 μW at the start of the measurements. It is not known if the quite large variations in the results for each sample reflects the sample or something to do with the measurements (vapor leakage, baseline variations etc.). After the samples were placed in the different climates from 54 to 85% RH all thermal powers decrease except for 85% RH. All samples with RH from 60 to 75% behaved rather similarly, but the samples at the lowest RH 54% showed lower activity after 40 days exposure. This is as expected as 54% RH is too low for HDM long term survival, whereas 60% RH is above the threshold for their extraction of moisture from the air at 20°C.

It is clearly seen that all activities decrease to very low values at the end of the exposure (76 days) in all ampoules, even those at 75 and 85% RH. This is a clear indication of that the way the HDM were exposed in this experiment was not optimal. It may be that the food source was not sufficiently nutritious or that some other factor prevented their continued growth. This must be improved before further measurements are made.

The blank samples with dog food showed low values except the ampoule at 85% RH and constant 20°C that increased to 101 μW after the high humidity exposure. This thermal power was generated by mold that was clearly seen in this ampoule (not seen in any other ampoules; but there may also have been some mold activity in the blank sample at 85% RH with varying temperature). The spread in the result from these blank samples is higher than expected. The values seems to be mainly close to zero or negative. This indicates that the ampoules may have been leaking water vapor as vaporization is one of the few processes that gives negative thermal powers.

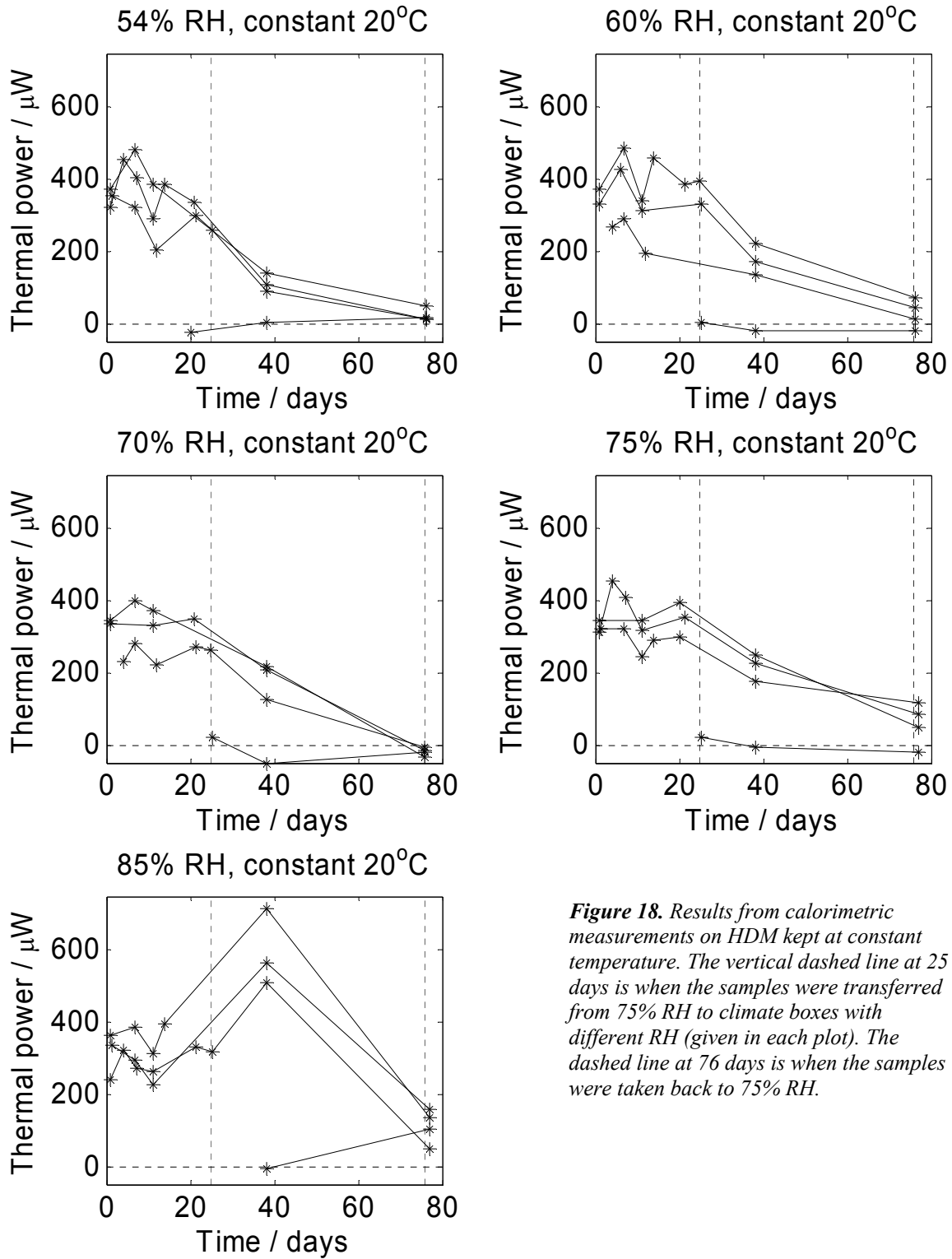


Figure 18. Results from calorimetric measurements on HDM kept at constant temperature. The vertical dashed line at 25 days is when the samples were transferred from 75% RH to climate boxes with different RH (given in each plot). The dashed line at 76 days is when the samples were taken back to 75% RH.

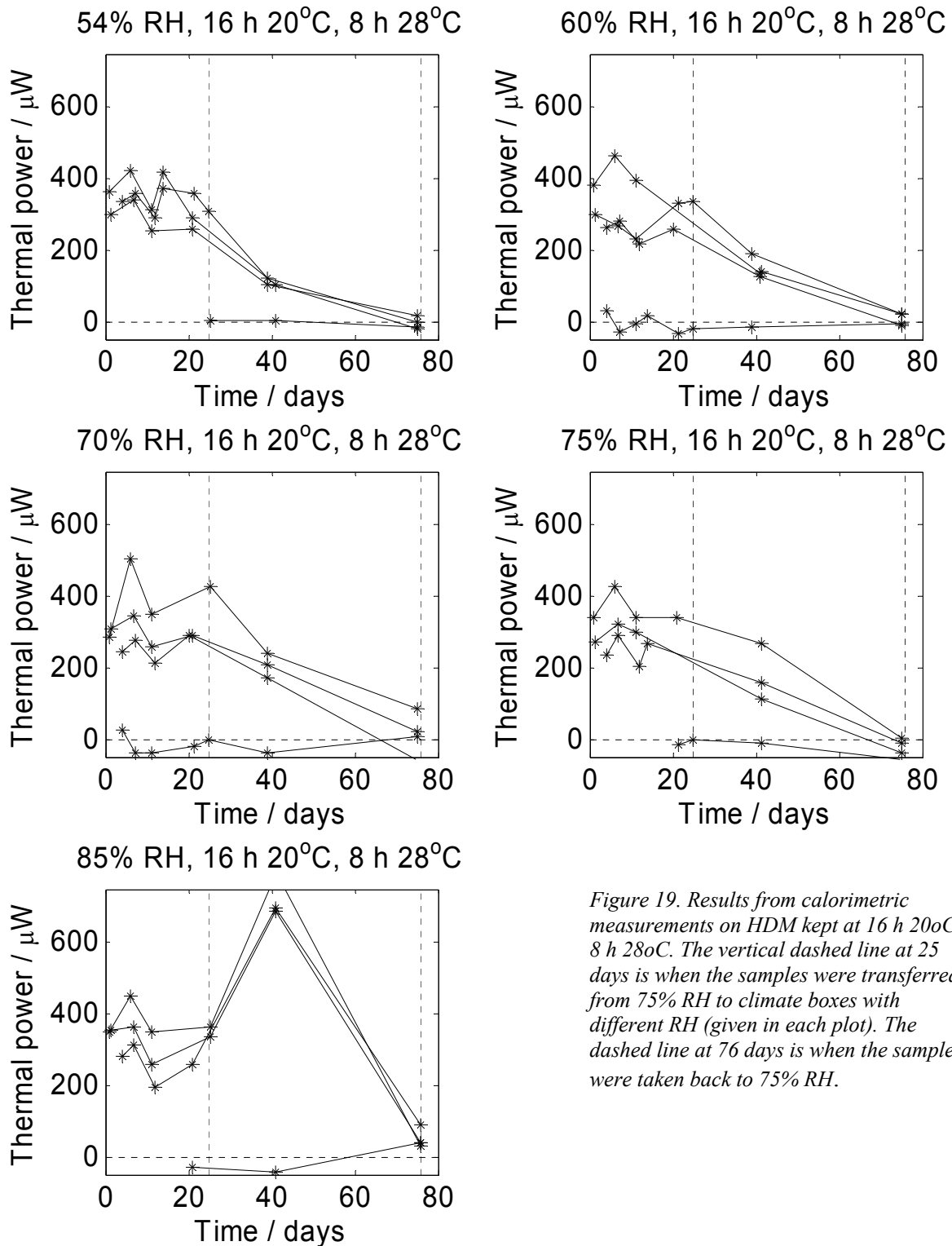


Figure 19. Results from calorimetric measurements on HDM kept at 16 h 20°C, 8 h 28°C. The vertical dashed line at 25 days is when the samples were transferred from 75% RH to climate boxes with different RH (given in each plot). The dashed line at 76 days is when the samples were taken back to 75% RH.

Petri dish method of measuring HDM activity

Method

Each 50 mm plastic Petri dish was charged with 50 HDM and 100 mg dog food. The dishes were then exposed to the following climates and measurements:

- 21 days at about 20°C and 75% RH.
- Counting (C1).
- 41 days at the ten climates of the climate boxes.
- Counting (C2).

Results

The result of the HDM counting in the Petri dishes is shown in Table 3 and in Fig. 20. The results for the varying climate (16 h at 20°C and 8 h at 28°C) are as expected with higher activity at higher humidities, except at 85% at which the activity is slightly lower than at 75%. This is usually attributed to competition from molds at higher RH, and the food particles in the Petri dishes at 85% RH were moldy. At constant temperature the picture is not at all as clear. The lowest counts are seen at 70% RH and quite high activity is seen also at 54% RH.

Table 3. Results from counting HDM in Petri dishes. Count C1 was made after 21 days at optimal climate 75% RH and about 20°C. Count C2 was made after 41 days exposure at the climates indicated in the leftmost column. The two values given refer to the two Petri dishes used for each exposure. Only moving HDM are counted. Both at count C1 and count C2 were HDM of different sizes and eggs seen. The counted values are rather approximate as some HDM may hide under the dog food.

Constant 20°C				
RH	at start	count C1	count C2	comments
54%	50; 50	50; 50	95; 150	
60%	50; 50	65; 50	120; 190	
70%	50;50	50; 50	45; 110	
75%	50; 50	50; 50	100; 120	
85%	50; 50	50; 50	105; 125	moldy
16 h at 20°C and 8 h at 28°C				
RH	at start	count C1	count C2	
54%	50; 50	50; 50	45; 55	
60%	50; 50	50; 50	55; 100	
70%	50; 50	50; 50	100; 135	
75%	50; 50	50; 50	140;150	
85%	50; 50	65; 50	115; 160	

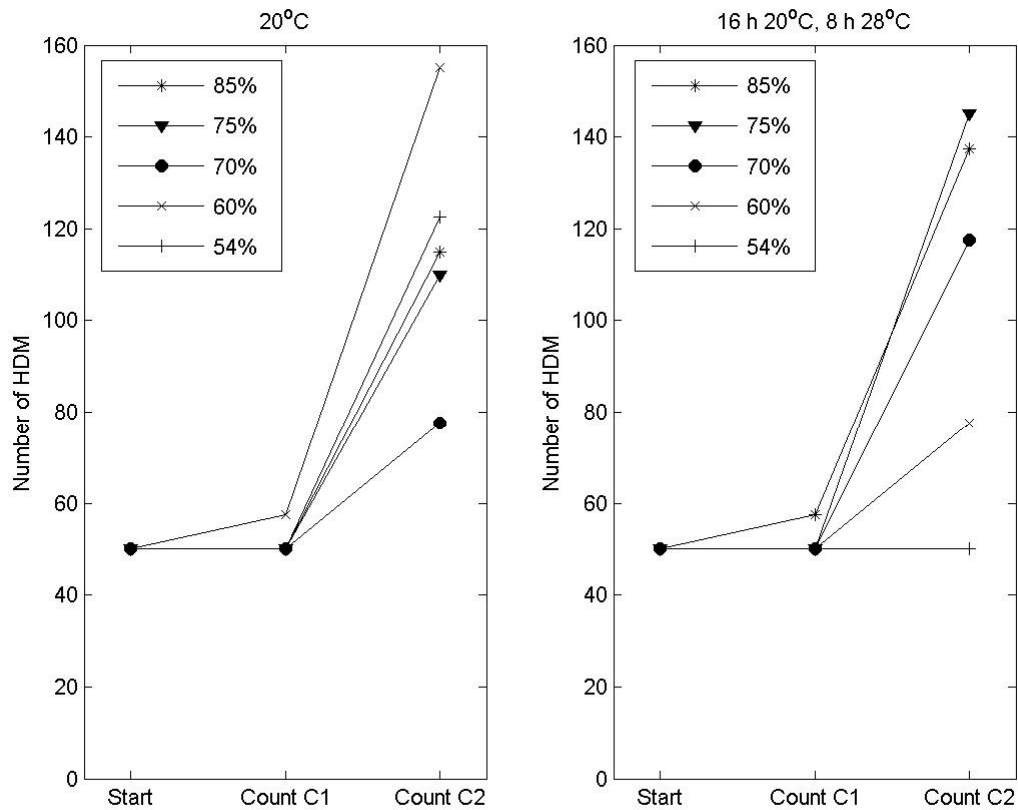


Figure 20. Results from Petri dish counts. Each point is the mean from counts of the two dishes in each climate.

The unexpected results for the constant climate conditions was maybe caused by the rather long initial exposure time at ideal conditions during which the HDM had laid eggs that were later hatched. The quite different number of individuals counted in some dishes from the same climates may be an indication of this.

Image analysis to find moving HDM in Petri dishes

We have also photographed Petri dishes with HDM to see if we could detect moving HDM. Images were taken with 5 s intervals with a macro objective on a high resolution digital camera. The images were then subtracted as all objects that have not moved will then be removed. Figures 21 and 22 show an example where the method was successful and each moving HDM is seen as two black marks (initial and final position). From such a differential image it is possible to calculate both size, speed and direction of each HDM on a two-dimensional surface. Figures 23 and 24 show a case where probably the camera moved between the exposures (the camera was fastened to a stand, but it was operated manually), so that also non-moving objects are seen. However, this should not be a problem if the camera is operated via cable.

The image analysis was done with the Image Analysis Toolbox in MATLAB 7.0.1 with the following steps (steps in brackets were not done in the present study, but could quite easily be implemented):

- form the difference between the two pictures
- gray-threshold and make black and white (objects that have moved are black)
- remove objects less than, e.g., 30 pixels
- fill in any holes in black areas
- count the number of objects
- (Remove all objects outside of Petri dish)
- (Remove all object that do not have the roundness factor that HDM have; such objects are likely food particles that have moved)
- (Find all objects)
- (Group objects in pairs so that similar objects at close distance are grouped together = one HDM)
- (Calculate size (area) of each HDM)
- (Calculate speed and distance)

One problem with the image analysis is that one can only count HDM that are moving; one does not count HDM that are attached to food particles. This problem may be avoided by the use of either a food covering the whole dish or by using very small food particles.

Image analysis could be a rational way of assessing the status of HDM, and maybe even to count how many HDM of different sizes there are.

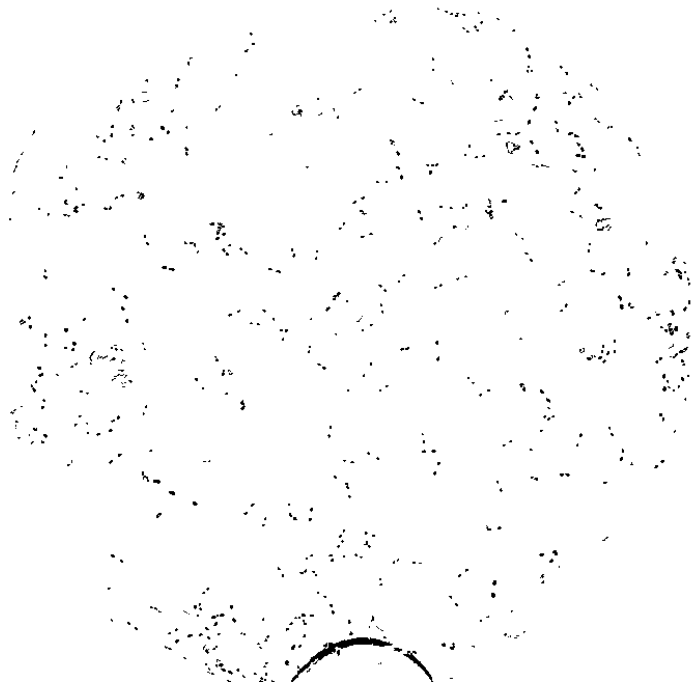


Figure 21. Successful differential image of HDM moving on a Petri dish. Each moving HDM is represented by two dots.



Figure 22. Zoom in on an area in Fig. 21 showing that each HDM is seen as two dots and that different HDM have different size..

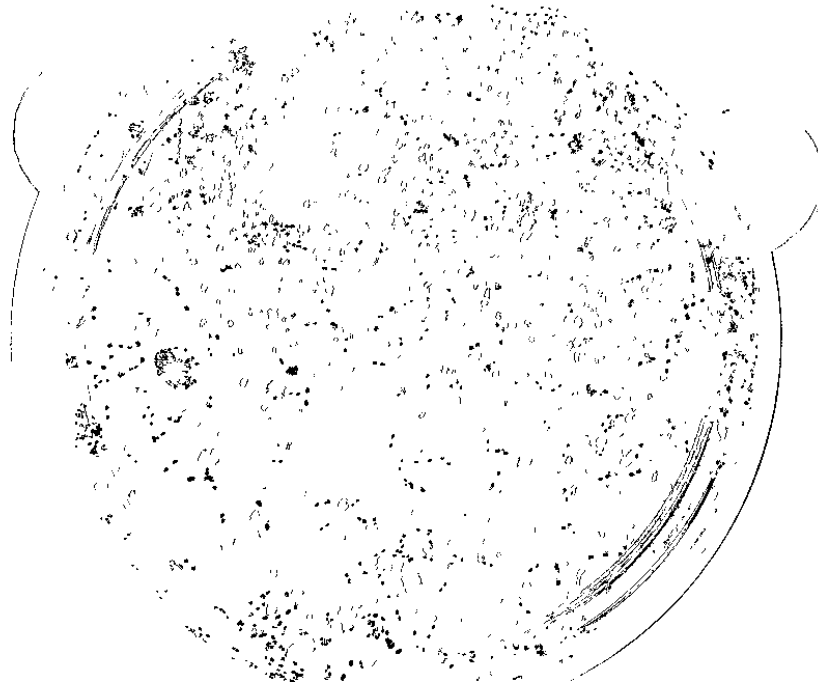


Figure 23. Not so successful differential image; probably because the camera moved between the exposures.



Figure 24. Zoom in on an area in Fig. 23. Moving HDM are seen as two dots, but there are also other objects (food particles, dead HDM) showing up as objects with lower degree of roundness.

4. Field measurements of microclimates in beds

The objective of this study was two-folded: to confirm the usefulness of a novel measuring equipment for microclimatic measurements in the indoor environment and to increase the knowledge of the temperature and moisture conditions in beds during use. This report presents the characteristics of the measuring equipment and the diurnal temperature and relative humidity variations in mattresses and bedding under normal use that have been measured for two different mattress types.

Measuring equipment

The requirements for the measuring equipment was that it should not impose discomfort for the persons sleeping in the beds and it should be robust enough to stand friction from moving persons. It should also be relatively inexpensive as a large number of sensors are needed to map the climate in a bed. To meet these requirements a combined temperature and relative humidity sensor connected to a logger device that can be operated in a “stand-alone”- mode was made in the electronics workshop at the School of Civil Engineering, Lund Institute of Technology. Each part of the measuring equipment is described below.

The combined sensor (se Fig. 24) was made from a commercially available RH-sensor and a thermistor. The RH sensor type has previously been used by (Baker, 2004 #43). (Cunningham, 1999 #121) have earlier used shrink tube to protect RH-sensors from dirt and water. In the shrink tube we made a small hole to keep the moisture response time of the sensor at a reasonable level.

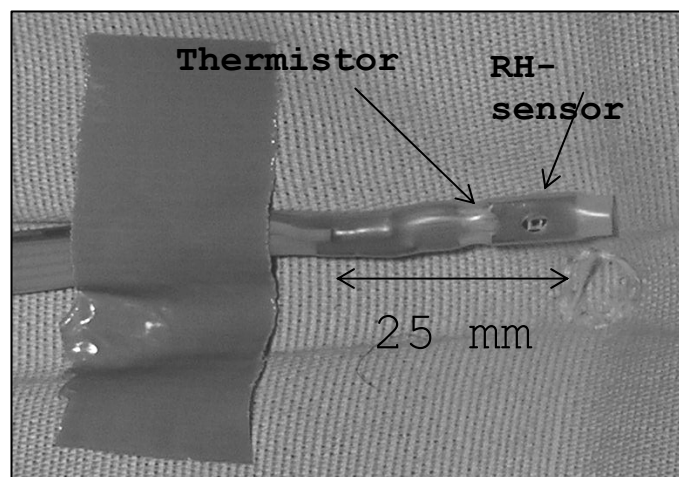


Figure 24. The combined temperature and RH sensor mounted on a mattress.

Thermistor

Precision resistance-temperature matched thermistors have been used. The accuracy for temperature measurement is $\pm 0.2^{\circ}\text{C}$ over the 0°C to 70°C temperature range. The time constant is 10 s in air.

Relative Humidity sensor

The relative humidity sensor (Honeywell HI-3610) is a commercially available polymer capacitive sensor with an on-chip integrated signal conditioning. The sensor has an accuracy of $\pm 2\% \text{RH}$ for the complete non-condensing interval and a maximum hysteresis of $\pm 1.2\%$

RH. The time constant in slow moving air is 15 s. Extended exposures at 90 % RH or higher causes a reversible shift of 3% RH, which have not been applicable for this study since the exposures have not been at so high level for extended time.

Logger

The logger (HgmLogger) simultaneously measures resistances (500Ω - $10 \text{ M}\Omega$) from eight thermistors and voltages (0-5 V) from eight RH-sensors. Each logger thus has 16 channels and can store 64000 data values. The logger can be used connected to a PC as well as in “stand-alone” mode.

Measurement set-up

In this field study two beds in two bedrooms in the same house were used for measurement of diurnal temperature and relative humidity variations. Both rooms are placed on the second floor and have the same orientation. The basic furniture is the same in both rooms, only the bed type differs, see the section “Mattress and bedding materials” below. A schematic drawing of room lay-out is shown in Fig. 25. The area and volume of each room is 10 m^2 and 25 m^3 respectively.

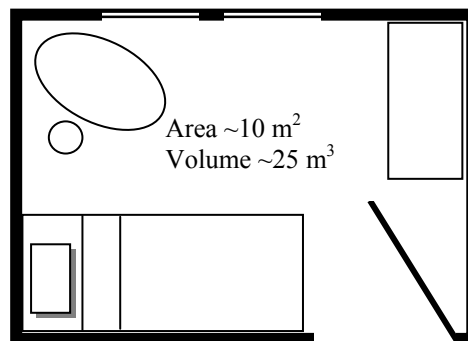


Figure 25. A schematic drawing of the room lay-out for Room 1 and Room 2.

Placing of sensors

In each measurement series 8 combined relative humidity and temperature sensors, as described above, were used. One sensor was placed in the center of the room approximately 2 m above the floor (sensor 8), one sensor was placed underneath the lath support in the bed (sensor 7) and 6 sensors were placed in the bed (sensors 1-6) as shown in Fig. 26. In the evaluation it was found that the RH sensor in sensor # 4 had a permanent failure. These results are omitted in the results presented in this report.

Occupancy

The beds were occupied by two young persons – a female for Bed 1 and a male for Bed 2. They are of the same body size and have approximately the same body mass. Time zero was set at the start of the first sleeping period in the study, approximately at 9 pm. Each sleeping period lasted for 9 – 10 hours. In this study there were no measurements made to take into account the differences in sleeping pattern and individual moisture and temperature production.

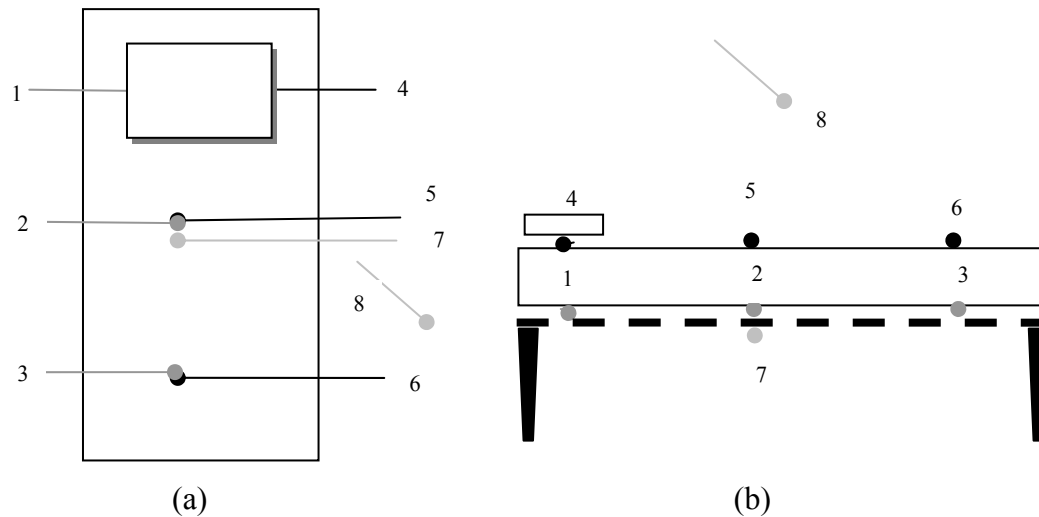


Figure 26. The placement of the sensors are shown from above (a) and from the side (b). Note that for Bed 1 the mattress studied is on top of a interior sprung mattress and sensors 1, 2 and 3 are not in contact with the wooden laths.

Mattress and bedding materials

Bed 1

The bed is an interior sprung mattress with an overlay mattress. The sprung mattress is supported by a bedstead with 70 x 19 mm wooden laths (60 mm spacing). The overlay mattress has a 40 mm polyether foam core which is covered on both sides with a 15 mm wool batting. The duvet has a helically crimped polyester fiber batting with a polyester/cotton cover fabric. The pillow is filled with polyester hollow fiber balls and also has a polyester/cotton cover fabric. The bottom sheet, duvet cover and pillow case are made of 100% cotton plain weave fabric. The space (260 mm high) underneath the bed was empty.

Bed 2

The bed is a 120 mm polyether foam mattress with a 100% cotton cover. The mattress is placed in a bedstead with wooden laths (66 x 19 mm with 60 mm spacing). The other bedding is identical to the one used in Bed 1. The space (270 mm high) underneath the bed was occupied with plastic boxes.

Results and discussion

Room climate

The climatic conditions (relative humidity and temperature) of the rooms have corresponding diurnal variations. Bed 1 shows a good agreement between the climate under the bed and in the room center. Bed 2 shows a higher RH and a lower temperature under the bed compared to the room center. The vapor content diagram shows that these difference is due to the temperature dependency of the relative humidity (Fig. 27).

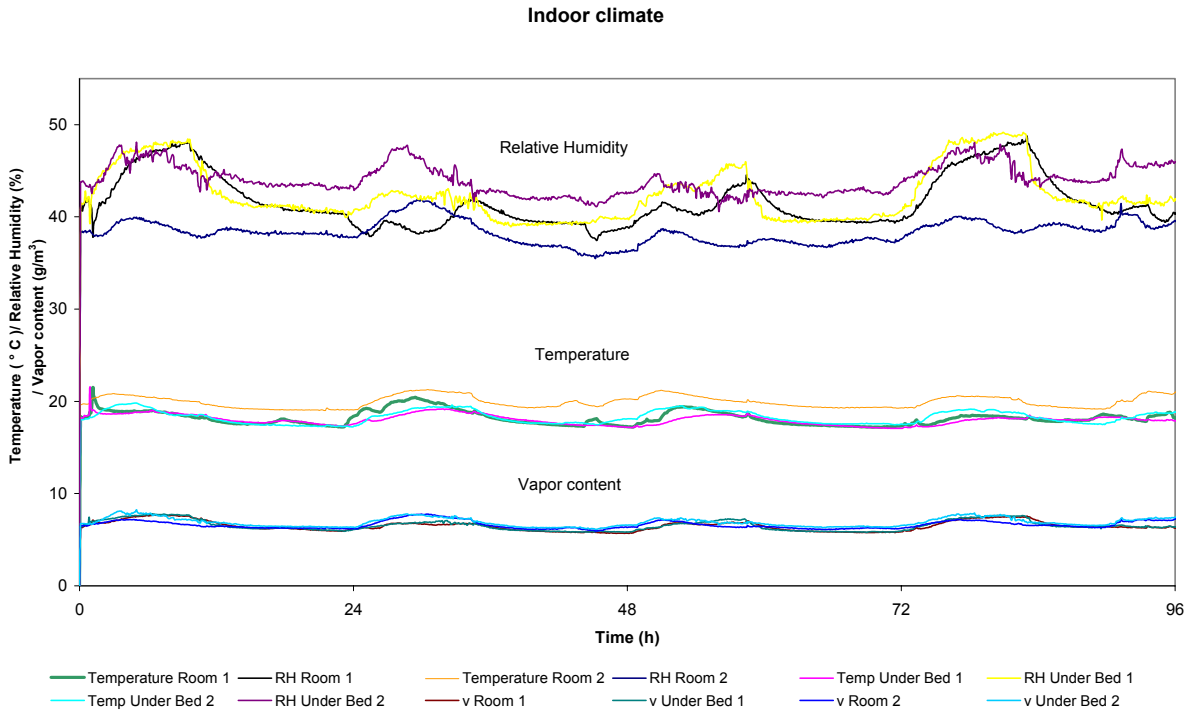


Figure 27. The indoor climate for Room 1 and 2 during the measurement period.

Temperature

The temperatures for Torso-top and Leg-top are the essentially the same for Bed 1 and 2, see Figs. 28 and 29. The differences seen in the head region may be due to individual sleeping patterns.

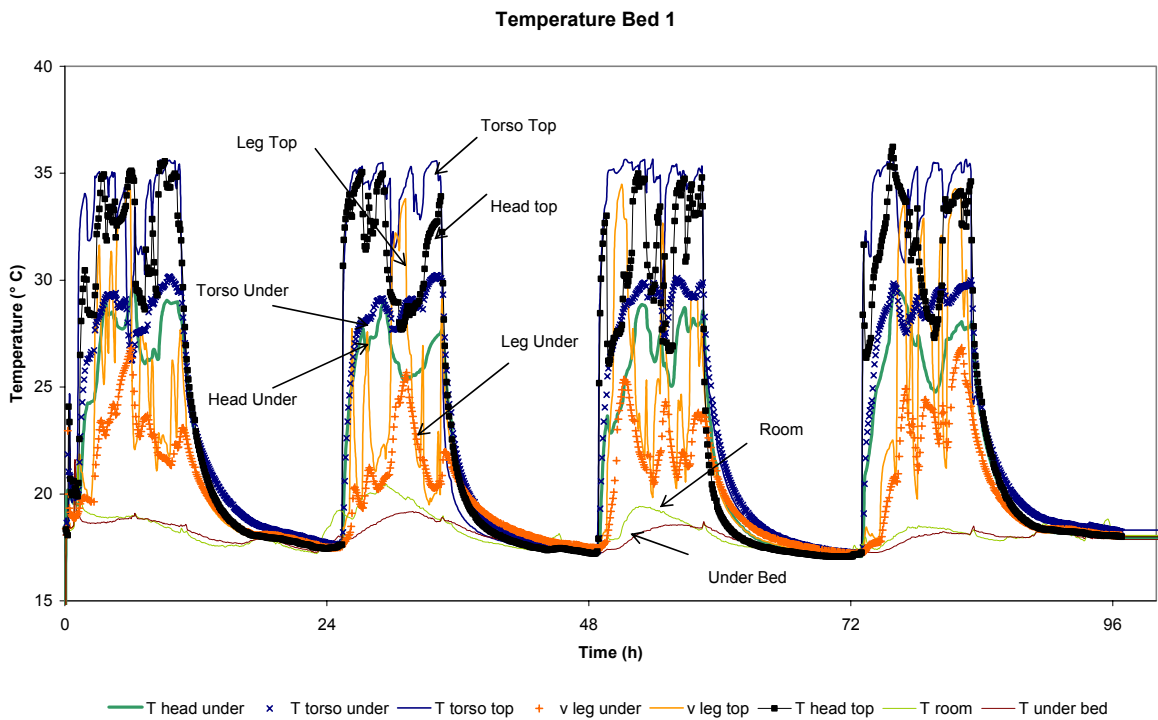


Figure 28. Temperature variations for Bed 1.

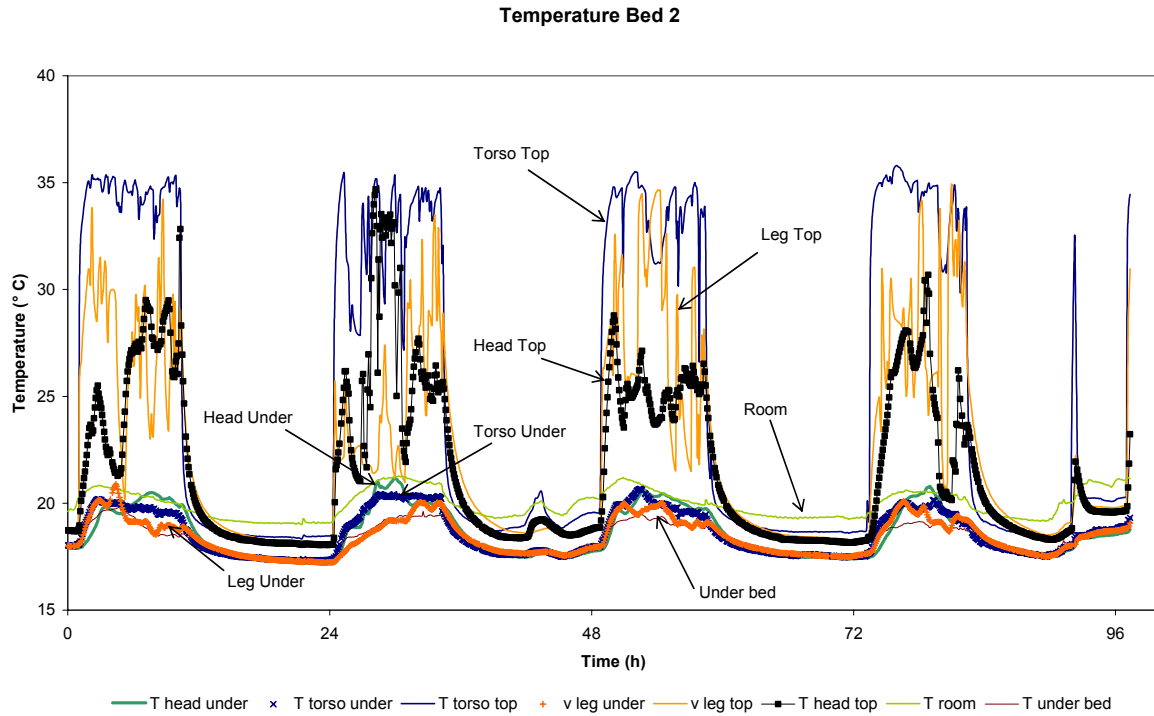


Figure 29. Temperature variations for Bed 2.

The temperatures measured under the mattress is lower for Bed 2 compared with Bed 1. This was expected as the thickness of the mattress in Bed 1 is only 58% of the mattress thickness in Bed 2.

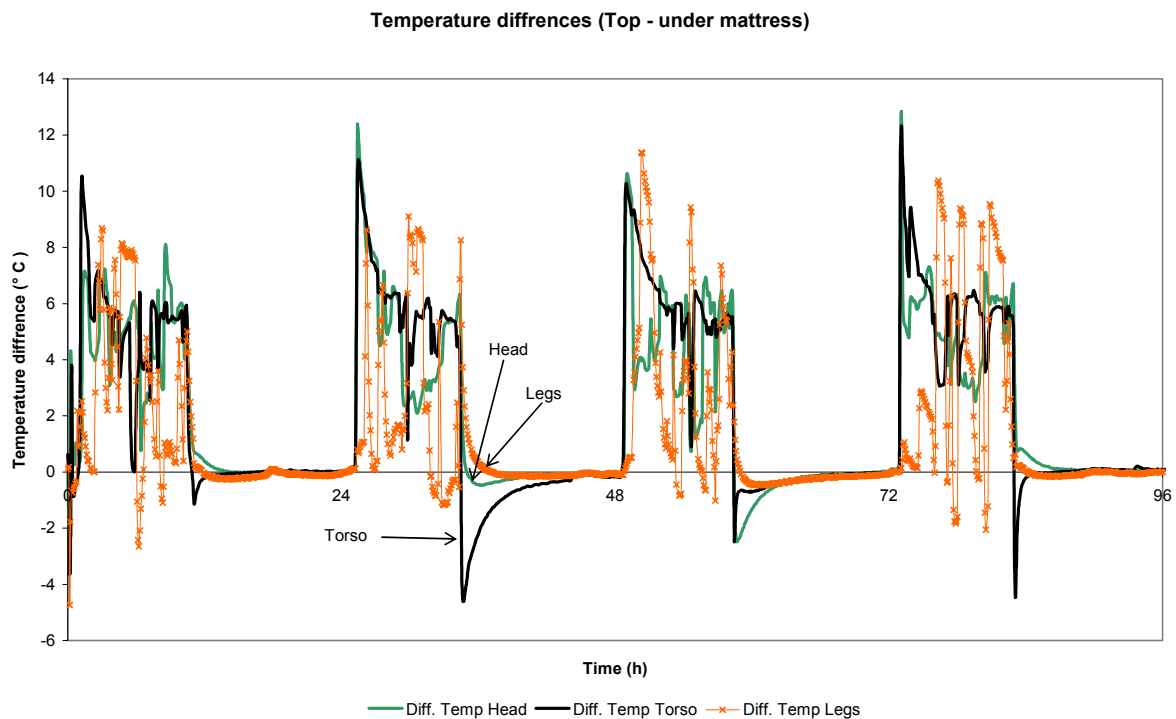


Figure 30. The temperature differences over the mattress in Bed 1 at three locations in the bed – head region, torso region and leg region.

In Fig. 30 the insulation effect of the underlying spring mattress in Bed 1 is shown. This can be seen in the negative temperature differences at the end of each sleeping period, due the

slower cooling under the mattress in Bed 1. This pattern is not seen for the mattress in Bed 2 (Fig. 31) which is exposed more directly to the room temperature on the bottom side of the mattress.

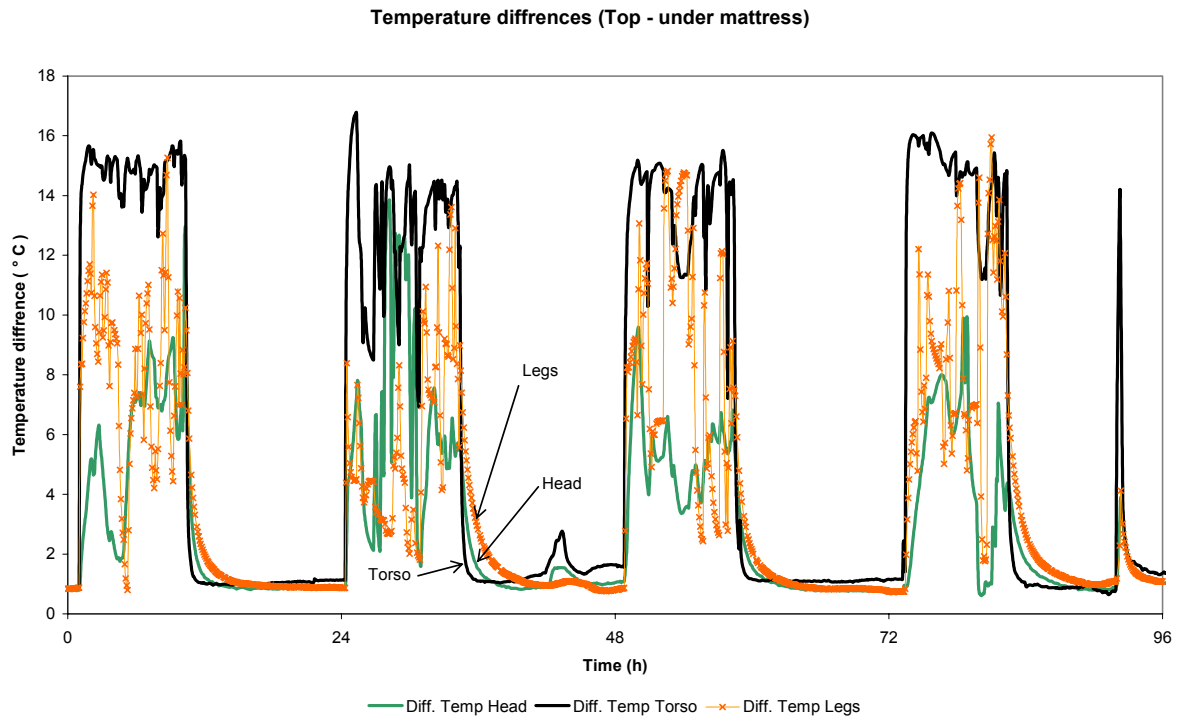


Figure 31. The temperature differences over the mattress in Bed 2 at three locations in the bed – head region, torso region and leg region

The effect of making the bed and putting the bed cover on is seen in Fig. 32. The morning after the second sleeping period Bed 1 was not made, it then follows the same cooling pattern as Bed 2 (never made during the measuring period).

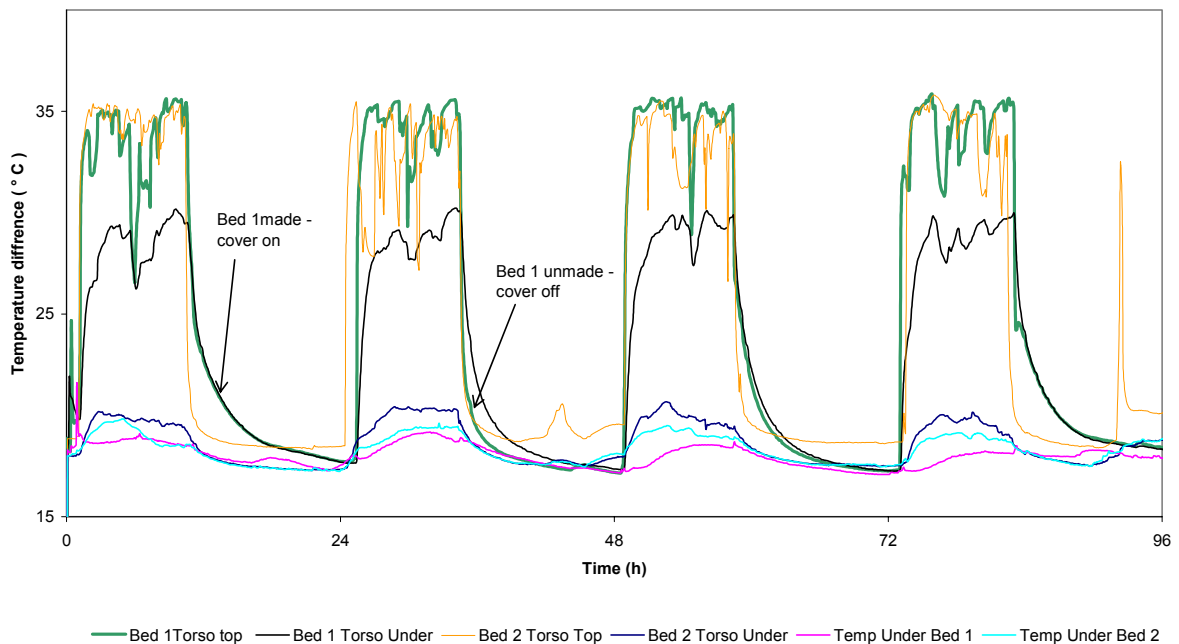


Figure 32. Temperature comparison between Bed 1 och Bed 2 – for the torso region.

Relative humidity

The results from the relative humidity (RH) measurements are hard to interpret, mainly because of the large fluctuations. These fluctuations are most probably due to the movement of the sleeping persons. Both beds show lower RH than expected, this is most evident for Bed 1 (Fig. 33) that barely reaches the 58% RH believed to be the lowest critical RH for house dust mite growth (CEH). The RH variations for Bed 2 also shows signs of moisture redistribution (Fig. 34).

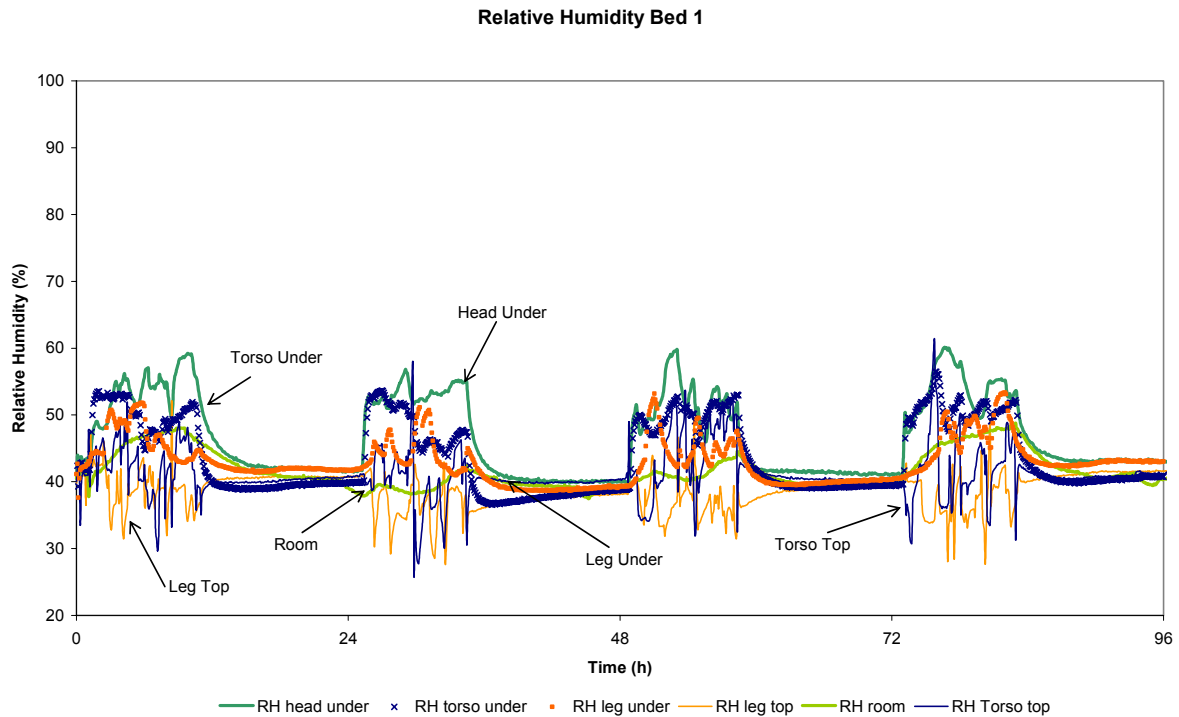


Figure 33. The variation in relative humidity for Bed 1.

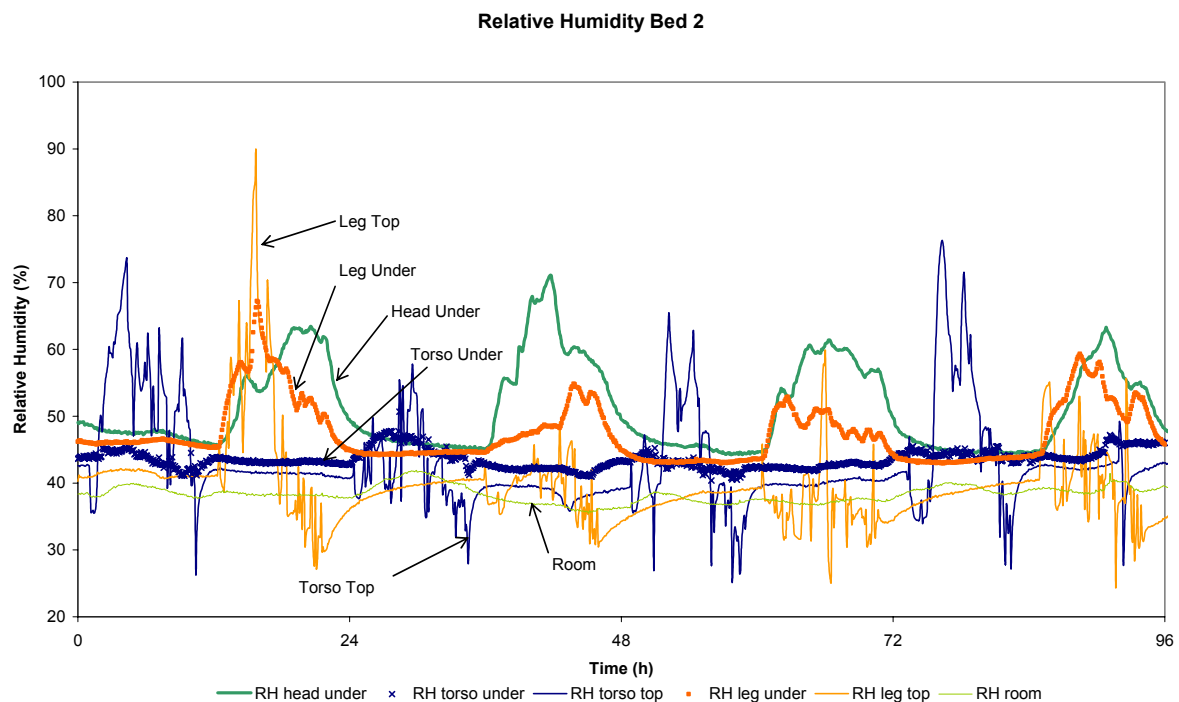


Figure 34. The variation in relative humidity for Bed 2.

Vapor content

The vapor content variations during the sleeping period are similar for both beds if the top of the mattress is considered. As expected, the bottom of the mattress in Bed 2 show less variations and follow the vapor content variations of the room. As for the relative humidity the vapor content results for Bed 2 suggests that we have a moisture redistribution during day-time which is not found for Bed 1 where the vapor content for both top and bottom of the mattress is approaching the room vapor conditions.

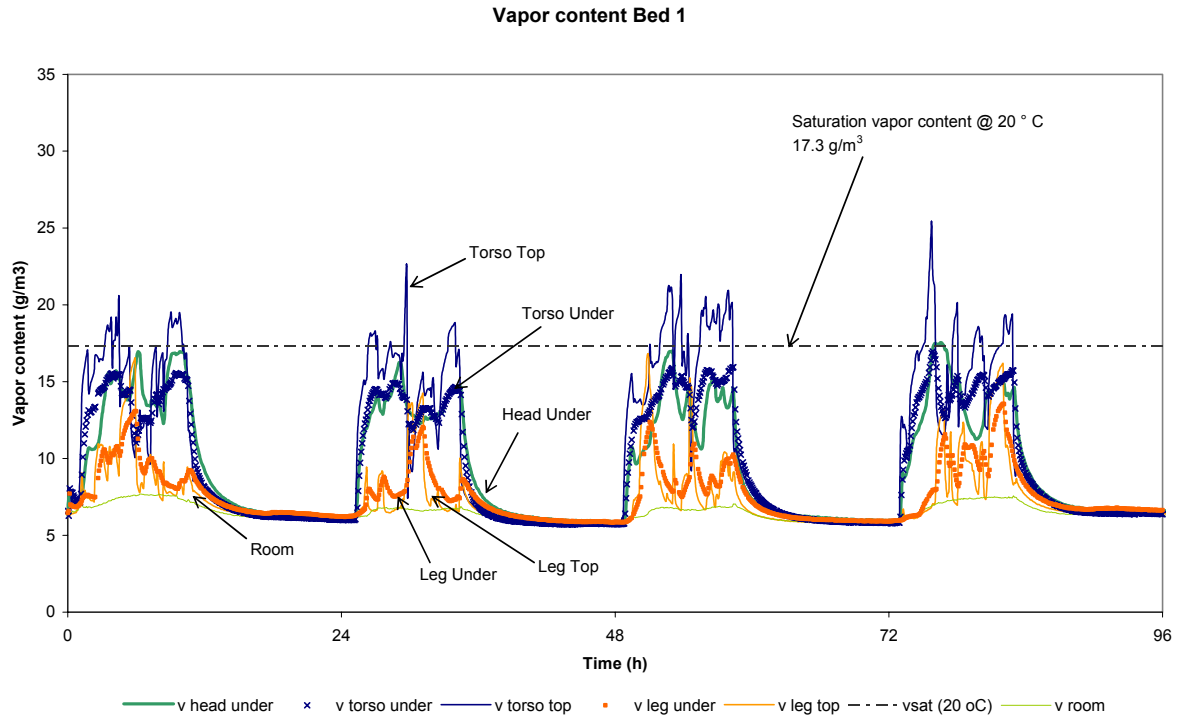


Figure 35. The variation in vapor content for Bed 1.

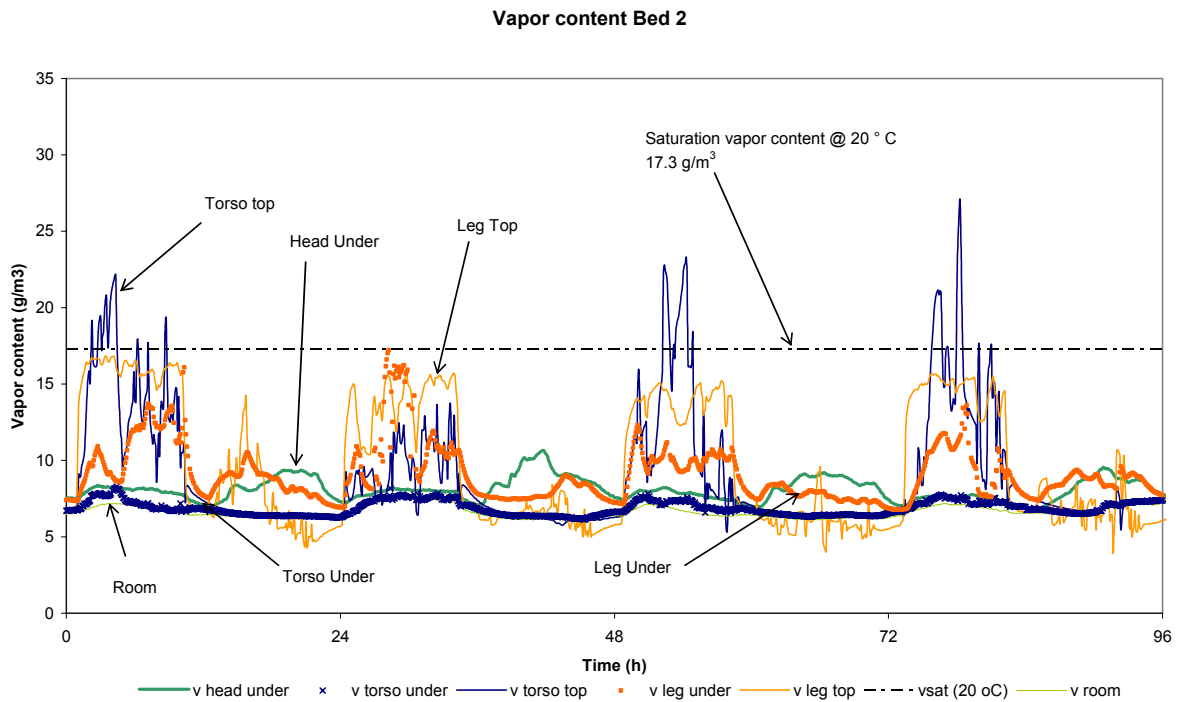


Figure 36. The variation in vapor content for Bed 2.

Also for the vapor content the impact of not making the bed is seen for Bed 1 at the end of sleeping period 2, the bed approaches the room vapor conditions quicker when left unmade and without bed cover, see Fig. 37.

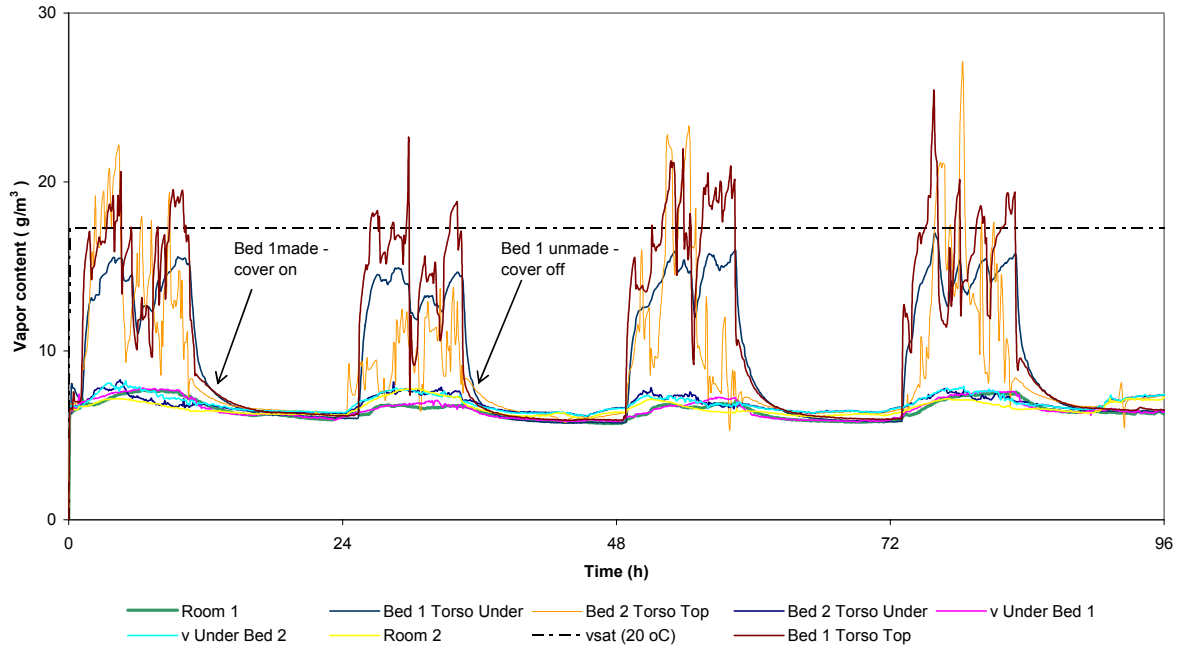


Figure 37. Vapor content comparison between Bed 1 and Bed 2 - for the torso region.

The equipment used to measure temperature and RH in beds worked well and did not give the sleeping persons any discomfort during the nights. The results from the bed measurements indicate that bed systems with different material combinations give noticeable differences in the measured microclimatic conditions for the beds during and between use. Further investigations regarding the importance of sleeping pattern and individual moisture production needs to be done. The temperature during the sleeping period increased significantly directly under the sleeping person while the relative humidity had a more modest increase.

5. Discussion

We have made a literature study on HDM and relevant indoor microenvironments. One obvious conclusion from this is that quite a lot of work has been done in the field, but there are still a number of questions that needs further research (cf. (Cunningham, 1996; Pretlove et al., 2001). One such question regards the aim of HDM control measures: should the aim be to have zero HDM or to minimize the HDM population? Although an RH continuously below 50% will eradicate an HDM population, many researchers propose instead to aim for low populations and low allergen levels (de Boer, 1998) as it is unrealistic to keep such low RH in *all* parts of a building, including beds, sofas etc.

How shall one model the influence of temperature and humidity on an animal with a complex life cycle with five stages? Should one find what is needed to prevent one stage from survival and thus hinder the life cycle to be completed, or does one have to take all stages into consideration? Literature indicates that there are differences between Df and Dp , but the differences may be so small that it is irrelevant to make a difference between them.

HDM can move rather rapidly. On a flat surface they can walk a distance in the order of their body length in 1-2 s. It is also clear that they have means of assessing their present environment (temperature and humidity), so that they will stay where the microclimate is good for them. However, it is not known how well they can search for better environments, for example by sensing temperature and RH gradients. Concerning, e.g., mattresses it is a critical question whether HDM can move within a mattress and if this movement is significant. Can HDM have daily movements to always find the optimum RH and temperature in a bed? HDM movement is investigated in a project in the UK (Pretlove et al., 2001), but it seems rather difficult to include such behavior into, e.g., a bed model. Our image analysis technique may be a method for such studies.

How detailed does a room or bed model have to be? Can one work with a one dimensional cross section of a bed (maybe a worst case profile), or does one have to model the bed as three dimensional? The latter is much more difficult, but may be necessary if HDM movements are significant.

Of more fundamental character is the question of what the salt solution used for the active uptake consists of. Chemical analysis seems to give a result incompatible with observed CEH.

Concerning our own studies it seems that the food used for the HDM needs to be improved. The decrease in HDM activity after the first 30-40 days indicate that the nutrient source was not good enough. It is possible that it contained a high percentage of indigestible material.

The calorimetric large scale measurement technique was successful, but needs some improvement to increase through-put. First of all is it necessary to design a new way to seal the ampoules so that no HDM can escape and no vaporization takes place during the calorimetric measurements. The seal should also be quick to apply and remove. Secondly the calorimeter needs to be improved concerning number of channels, sensitivity and baseline stability. None of the above points should pose any major problems.

It is known that the optimal climate for HDM population development is around 25°C and 75% RH. At higher temperatures they are probably limited by difficulties in keeping a water balance and at higher humidities they cannot successfully compete against molds and predators. According to our studies HDM die after 45°C for 30 minutes and this could possibly be a way to control HDM populations. One could for example regularly heat beds above this temperature. However, a difficulty with this is that HDM try to escape harmful

temperatures, so – unless the whole bed is treated – they may emigrate to other parts of a bed during the treatment and come back later.

The idea of using a Millipore filter to stop HDM from leaving the ampoules, but still allow gas and vapor exchange, was successful. However, it would be an advantage if the mass transfer rate between ampoule content and ambient conditions could be increased, at least by a factor of 2. This may be possible by use of another seal technique that reduces the distance between sample and Millipore filter.

The material properties for the bedding materials and mattresses need to be determined. The foam and batting materials are compressed during use so how to determine material properties for these compressible materials is a challenge. Properties that are needed are thermal conductivity, heat capacity, water vapor diffusivity, sorption isotherms, and possibly also a measure of how easy the HDM can move in the materials.

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Appendix A - Background studies

Here we summarize some background measurements and calculations for the calorimetric studies. It is often not trivial to work with moisture as, e.g., changes in moisture state may take longer time than expected. The studies described below both show that the present measurements have worked as expected, but also suggest some future improvements.

Moisture state of boxes

Ten plastic boxes with saturated salt solutions to generate different relative humidities were used in the present experiments. Five of them were placed in constant 20°C and five were placed in a climate cabinet with a program of 16 h 20°C and 8 h 28°C. In each climate box temperature and relative humidity were measured with a Hobo-logger. Table A1 shows an overview of the first phase of the experiments that lasted 36 days.

Table A1. Overview of climate boxes and results from measurements of the climate in the boxes. Subindex “lit” denotes literature values from Nyqvist [21]. Relative humidities (RH) are given as “%” and temperatures (T) are given in °C.

Constant 20°C							
Salt	RH _{lit}	RH _{mea}	T _{mea}	Comments			
Mg(NO ₃) ₂	54	53	21.0				
NaBr	59	57	21.0				
KI	70	69	21.0				
NaCl	75	74	21.3				
KCl	85	85	21.0				
16 h 20°C, 8 h 28°C							
Salt	20°C			28°C			Comments
	RH _{lit}	RH _{mea}	T _{mea}	RH _{lit}	RH _{mea}	T _{mea}	
Mg(NO ₃) ₂	54	52	19.8	52	49	27.9	
NaBr	59	58	19.8	57	55	27.9	salt dried out after 23 days
KI	70	69	19.8	68	67	27.9	salt dried out after 30 days
NaCl	75	74	19.8	75	73	27.9	salt dried out after 30 days
KCl	85	85	19.8	84	82	27.9	

The loggers were not calibrated or validated before these measurements. However, all loggers in the same temperature gave almost exactly the same result (± 0.1 K) and the loggers seems also to have been well calibrated ($\pm 2\%$ RH) for relative humidity measurements.

Measurements with loggers were made for about 35 days (result shown in Figs. A1-A2). Then it was discovered that some salt solutions had dried out and water was added to these, before the HDM exposures continued for another 14 days.

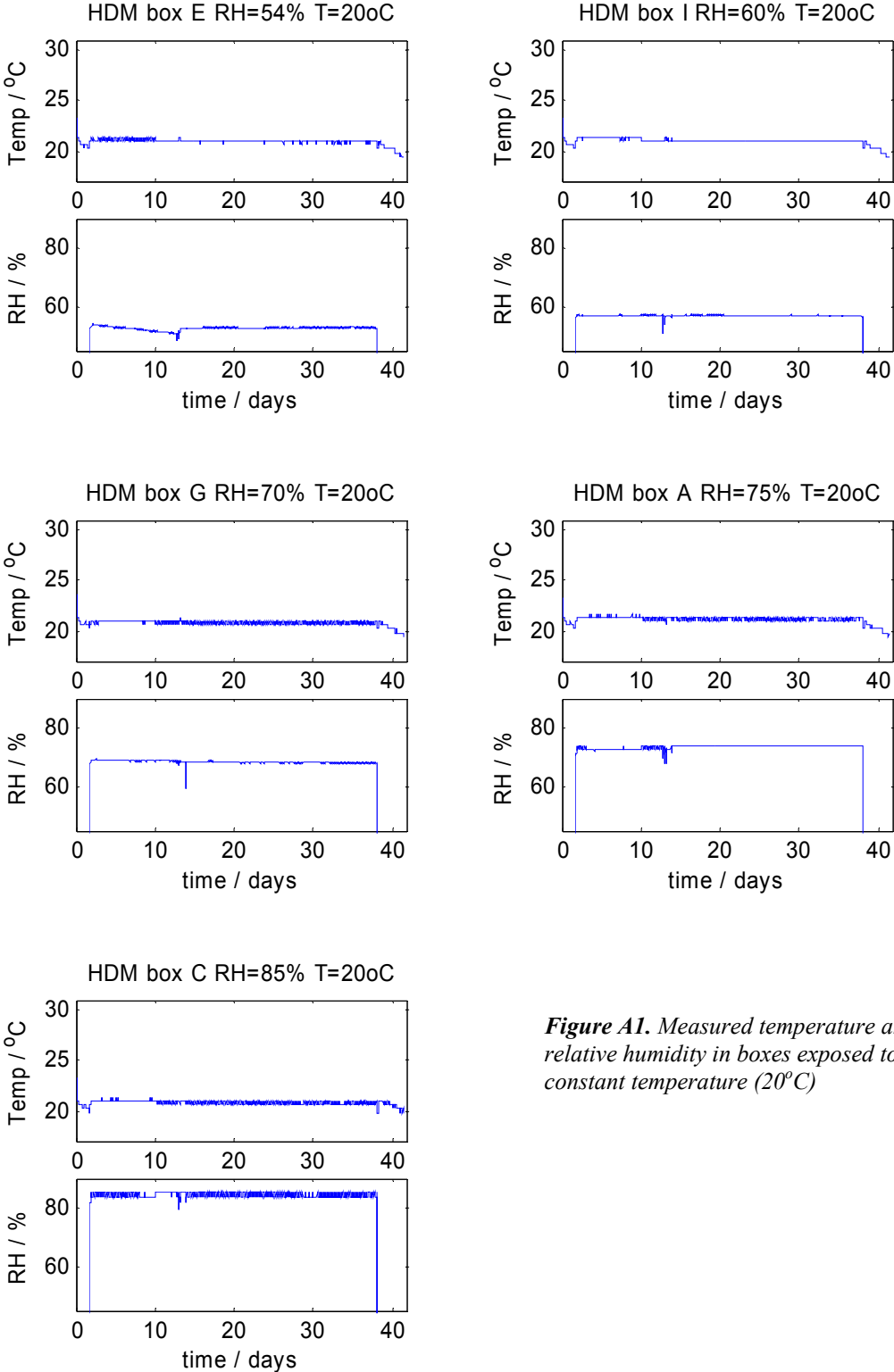


Figure A1. Measured temperature and relative humidity in boxes exposed to constant temperature (20°C)

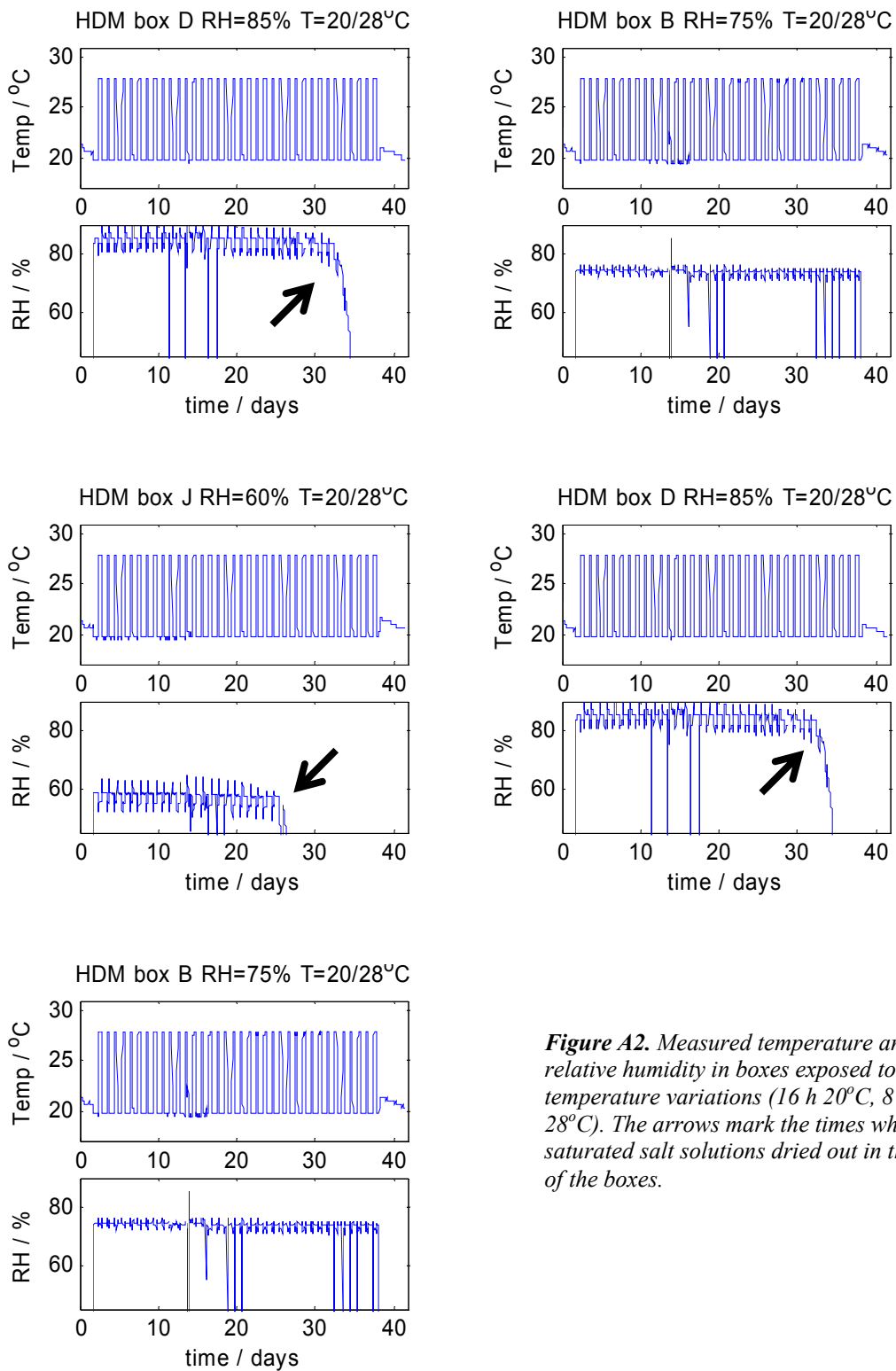


Figure A2. Measured temperature and relative humidity in boxes exposed to daily temperature variations (16 h 20°C, 8 h 28°C). The arrows mark the times when the saturated salt solutions dried out in three of the boxes.

Moisture state of sample

Above it was shown that all boxes with HDM samples have the expected relative humidities. However, this does not mean that the samples of dog food and HDM experience these expected relative humidities as there is a mass transfer resistance from the RH in a box to the sample and certain amounts of moisture needs to be transported to the sample in order to get the expected RH in the HDM environment. We have therefore made measurements and calculations to investigate this problem. The nomenclature used here is given below:

k	mass transfer coefficient	g/s/Pa
m	mass	g
m_0	dry mass	g
p	vapor pressure	Pa
p_{sat}	saturation vapor pressure	Pa
q_m	mass flow rate	g/s
ϕ	relative humidity (p/p_{sat})	1
ξ	moisture capacity*	1

* The moisture capacity of the mass of vapor absorbed per dry mass per change in relative humidity (i.e., slope of the sorption isotherm)

Mass transfer resistance from box to sample

Some ampoules were made with water (instead of HDM samples) and placed in 75% RH boxes together with the normal samples. The mass losses of these ampoules were used to assess the mass transfer resistance from box air to a sample. The approximate mass losses were 1 gram water per month, i.e. $4.1 \cdot 10^{-7}$ g/s. The mass transfer can be described by the following equation:

$$q_m = k \cdot \Delta p . \quad (A1)$$

At 20°C (at which most of the measurements took place) $p_{sat}=2340$ Pa. In the present case the relative humidity difference was only $1.00-0.75=0.25$ of that, so the mass transfer coefficient k is $4.1 \cdot 10^{-7}/0.25/2340 \approx 7 \cdot 10^{-10}$ g/s/Pa.

Water vapor sorption by sample

The HDM samples contains 500 mg dog food and 100 mg HDM. We have measured the sorption isotherm of the main component, the dog food, and take that as representative for the whole sample. The absorption and desorption isotherms were measured on a finely divided sample of about 60 mg in a DVS 1000 sorption microbalance. The result showed that the sorption rate was rather low (the sample material itself was slow in sorbing moisture). In the following calculations a mean of the absorption and desorption isotherms have been used.

A most important parameter in this type of calculations is the moisture capacity ξ , i.e. the change in moisture content per change in relative humidity:

$$\xi = \frac{d(m/m_0)}{d\phi} \quad (\text{A2})$$

Note that the relative humidity ϕ is given as a fraction. Figure A3 gives the moisture capacity calculated from the sorption measurements. It is seen that it is quite constant at about 0.15 at RH lower than 0.6, but that it sharply increases at high RH. A higher moisture capacity will mean that more moisture has to be transported, i.e. longer times. In the calculation below we have therefore used $\xi=1$ that is a relevant value for the highest humidity range 0.75-0.85 used in the present experiments.

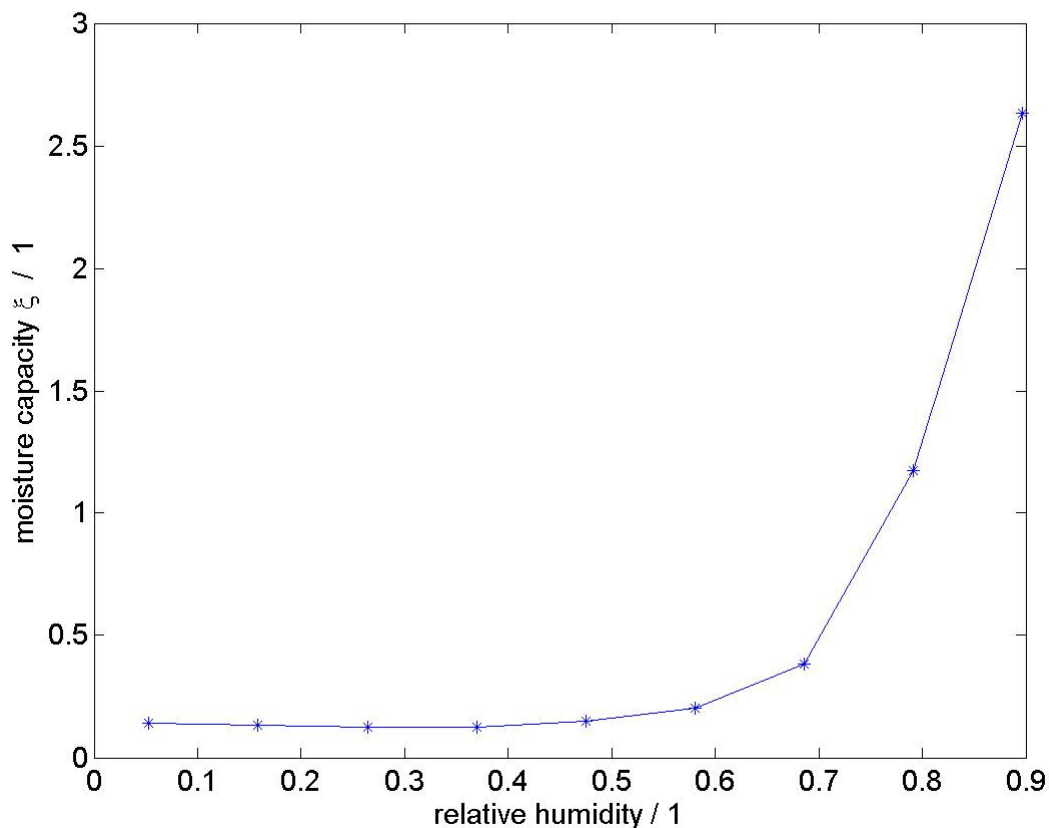


Figure A3. The moisture capacity of the dog food that was the major part of the sample.

A model for the mass transfer

We view the sample ampoule as a moisture capacity (the sample) connected to a constant RH (the box) through a mass transfer coefficient (the Millipore membrane and the air in the ampoule). It can then be shown that Eq. A1 above together with a mass balance gives the following solution to the how rapidly the relative humidity ϕ in a sample changes after a step change from ϕ_1 to ϕ_2 in the box:

$$\frac{\phi(t) - \phi_2}{\phi_1 - \phi_2} = \exp(-t / \tau), \quad (\text{A3})$$

where the time constant τ is defined as:

$$\tau = \frac{\xi \cdot m_0}{k \cdot p_{\text{sat}}}. \quad (\text{A4})$$

The time constant of the process increases if the sample moisture capacity (ξ) or size (m), increases, or if the mass transfer coefficient (k) or saturation vapor pressure (p_{sat}) decreases.

For the present (worst) case calculation, $\xi=1$, $m_0=0.5$ g, $k=7 \cdot 10^{-10}$ g/s/Pa, and $p_{\text{sat}}=2340$ Pa, which gives a time constant τ of $3 \cdot 10^5$ s, or about 4 days. This is the time during which 63% of a final change has taken place. After 3 time constants (about 11 days) 95% of a change has taken place.

Conclusion

The above calculations show that it does take some time for the sample to adjust to a new external RH, but that most of the changes have taken place within 10 days, so that long term (several weeks) measurements are made with the similar RH-conditions to those expected from measurements in the external climate. However, it would be advantageous to decrease the time constant, for example by decreasing the sample size or increasing the mass transfer coefficient.

The above calculations also show that the moisture state of the ampoules are not disturbed by handling the uncapped samples for a few minutes before they are capped or put back into the climate boxes.

Oxygen state in the ampoules during measurement

For the calorimetric measurements the ampoules have to be closed. The oxygen pressure will therefore decrease during a measurement. Can this influence a measurement made 120 minutes after the sample was charged into the calorimeter?

The oxygen pressure p (Pa) in the air is about 21000 Pa (21% of the atmospheric pressure). The amount n (mol) of oxygen in an ampoule can be calculated with the ideal gas law:

$$pV = nRT \quad (\text{A5})$$

Here, V (m^3) is the gas volume of the ampoule, R is the gas constant and T (K) is the temperature. As the gas volume of the ampoule is about 2.5 ml, the amount of oxygen is about $21000 \cdot 2.5 \cdot 10^{-6} / 8.3 / 293 = 22 \mu\text{mol O}_2$. All aerobic biological activity produced about 469 kJ per mol O_2 consumed. The highest thermal powers measured initially were about 400 μW . This gives that about $400 \cdot 10^{-6} / 469 \cdot 10^3 \cdot 120 \cdot 60$, that is about 6 $\mu\text{mol O}_2$ is consumed during the 120 minutes before a measurement is made. This is a significant decrease in oxygen pressure to about 73% of atmospheric oxygen pressure. It is not known how this affects HDM, but it may account for the slightly decreasing thermal powers seen during the measurements.