Exposure to trichloramine, trihalomethanes and endotoxins – adverse respiratory and ocular effects among Swedish indoor swimming pool workers

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ABSTRACT

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Indoor swimming pool facilities often use chlorine for pool water disinfection. Chlorine can also contribute to the formation of unwanted disinfection by-products (DBPs). Trichloramine and trihalomethanes (THMs) are DBPs formed in swimming pool water and swimming pool air causing occupational exposure. Trichloramine can cause ocular and airway irritation, while some THMs are potentially carcinogenic. Bacterial endotoxins may also cause adverse respiratory effects among swimming pool workers.

The overall aim of the study was to measure levels of trichloramine, THMs and endotoxins in the air at conventional, habilitation and rehabilitation, and adventure swimming pool facilities, and to investigate adverse ocular and respiratory effects among indoor swimming pool workers.

Trichloramine and THM levels in air were comparable to previous studies. Endotoxin levels in air were low compared to the reference value. Trichloramine concentrations in personal samples were approximately half as high as those measured in stationary samples - an important consideration for a future Swedish occupational exposure limit. The adventure facilities had the highest trichloramine levels while the conventional facilities had the highest THM levels. The adventure workers also reported more ocular and respiratory symptoms compared to referents (office workers) and had a lower FEV1/FVC ratio indicating airway obstruction. Workers both in conventional, and in habilitation and rehabilitation facilities exhibited a difference in FeNO levels compared to referents, implying airway inflammation.

The occupational exposure and adverse health effects found in indoor swimming pool environments emphasizes the importance of a personal based Swedish occupational exposure limit for trichloramine in air.

Keywords: trichloramine, trihalomethanes, endotoxins, occupational exposure, ocular health effects, respiratory health effects, swimming pool facilities

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SAMMANFATTNING PÅ SVENSKA

I svenska inomhusbad används ofta klor för desinfektion av bassängvatten. Klor har i regel god desinfekterande förmåga och dödar mikroorganismer, som annars kan sprida smitta. Kloret kan dock även bidra till att oönskade ämnen bildas. Exempel på det är trikloramin and trihalometaner som bildas i bassängvattnet och, som på grund av sin flyktighet, även avges till simhallsluften. Vid inandning av simhallsluften kan de anställda exponeras för dessa ämnen. Trikloramin är en förening som är känd för att orsaka irritation i ögon och luftvägar medan några av trihalometanerna är potentiellt cancerogena. I simhallarnas varma och fuktiga miljö kan även endotoxiner, som kommer ifrån bakterier, förekomma i luften. Endotoxiner utgör därmed ytterligare en yrkesrelaterad exponering i simhallsluft, som kan ha negativ påverkan på luftvägarna.

Det övergripande målet med denna avhandling var att mäta nivåer av trikloramin, trihalometaner och endotoxiner i luft i traditionella simhallar, habiliterings- och rehabiliteringsbad samt äventyrsbad och att undersöka negativa hälsoeffekter avseende ögon och luftvägar bland simhallspersonal.

Lufthalterna av trikloramin och trihalometaner, som uppmättes i det här arbetet, var jämförbara med de halter man uppmätt i tidigare studier. Trikloraminhalterna uppmätta med personburen mätutrustning på simhallspersonalen var ungefär hälften så höga som de som uppmättes stationärt runt om på baden – en viktig iakttagelse för ett framtida svenskt yrkeshygieniskt gränsvärde. Äventyrsbaden hade de högsta trikloraminhalterna medan de traditionella simhallarna hade de högsta trihalometanhalterna. Lufthalterna av endotoxiner var lägre jämfört med referensvärde för yrkesrelaterad exponering.

Simhallspersonalen i äventyrsbaden rapporterade också mer ögon- och luftvägssymtom jämfört med kontrollerna (kontorsarbetare) och hade tecken på luftvägsobstruktion, en sammandragning av luftvägarna som hindrar det fria luftflödet. Simhallspersonal i såväl traditionella som i habiliterings- och rehabiliteringsbad hade, jämfört med kontrollerna, inte samma minskning av kvävemonoxid i utandningsluft under arbetsdagen, vilket kan tyda på luftvägsinflammation.

I detta avhandlingsarbete påvisades yrkesrelaterad exponering och negativa hälsoeffekter bland simhallspersonal, vilket understryker vikten av ett svenskt yrkeshygieniskt gränsvärde för trikloramin i luft, som är baserat på personburen mätning.

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ABBREVIATIONS

ACGIH	American Conference of Governmental Industrial Hygienists
AM	arithmetic mean
ATD	automatic thermal desorption
BMI	body mass index
$Ca(ClO)_2$	calcium hypochlorite
CCl_3COOH	trichloroacetic acid
CH_2Cl_2N	dichloroacetonitrile
CH_3NCl_2	dichloromethylamine
$CHBr_3$	bromoform
$CHCl_2Br$	bromodichloromethane
$CHCl_2COOH$	dichloroacetic acid
$CHCl_3$	chloroform
$CHClBr_2$	dibromochloromethane
CI	confidence interval
Cl^-	chloride
Cl_2	chlorine gas
ClO^-	hypochlorite ions
CNCl	cyanogen chloride
COPD	chronic obstructive pulmonary disease
CV	coefficient of variance
DBPs	disinfection by-products
ECSC	European Coal and Steel Community
ELF	epithelial lining fluid
ERS	European Respiratory Society
EU	endotoxin units
FeNO	fraction of exhaled nitric oxide
FEV1	forced expiratory volume in 1 second
FNU	formazine nephelometric units
FVC	forced vital capacity
GC	gas chromatography

GM	geometric mean
GSD	geometric standard deviation
H_2O	water
HClO	hypochlorous acid
HMW	high molecular weight
IARC	International Agency for Research on Cancer
IC	ion chromatography
IgE	immunoglobulin E
IRR	incidence rate ratio
kU/L	kilo-units/liter
LAL	limulus amebocyte lysate
LC	liquid chromatography
LFT	lateral flow test
LLN	lower limit of normal
LMW	low molecular weight
LOD	limit of detection
LOQ	limit of quantification
LPS	lipopolysaccharides
LRT	lower respiratory tract
n	number of samples
NaClO	sodium hypochlorite
NCl_3	trichloramine
NEG	Nordic Expert Group for Criteria Documentation of Health Risks from Chemicals
NH_2Cl	monochloramine
NH_3	ammonia
$NHCl_2$	dichloramine
NO	nitric monoxide
OA	occupational asthma
OEL	occupational exposure limit
OH^-	hydroxide
OR	occupational rhinitis
OR	odds ratio

PEF	peak expiratory flow
ppb	parts per billion
PVC	polyvinylchloride
R^2	coefficient of determination
RADS	reactive airway dysfunction syndrome
RAST	radioallergosorbent assay
RUDS	reactive upper airway dysfunction syndrome
SD	standard deviation
SPSS	Statistical Package for Social Science
SWEA	Swedish Work Environment Authority
THMs	trihalomethanes
URT	upper respiratory tract
UV light	ultraviolet light
VC	vital capacity
VOC	volatile organic compounds
WEA	work exacerbated asthma
WER	work exacerbated rhinitis
WHO	World Health Organization
WMA	World Medical Association
X	independent variable
Y	dependent variable

INTRODUCTION

Sweden has just over 400 public indoor swimming pool facilities; most are operated by municipalities but some are privately run. Chlorination is the principal means by which pool water is disinfected to eliminate pathogens. However, although it is a good disinfector, chlorine helps form unwanted disinfection by-products (DBPs), some of which are known to cause adverse health effects among visitors and employees of indoor swimming pool facilities. Occupational exposure related to DBPs has been an recurrent issue for the specialists in the field of occupational and environmental medicine, as they regularly have treated patients who worked in indoor swimming pool facilities and suffered from work related respiratory symptoms.

Chloramines are DBPs that form in swimming pool water as a result of reactions between chlorine and nitrogen containing compounds. Due to its volatility, trichloramine is the most common chloramine in the ambient air of swimming pools where it is known to irritate eyes and airways. International studies have measured a wide range of trichloramine concentrations in swimming pool air and associated them with ocular and respiratory health effects.[1-4] The World Health Organization (WHO) was the first organization to set a recommended reference value for trichloramine exposure[5] but latterly, some European countries have established their own reference values for bathers.[6-9] Today's reference values are based on stationary sampling; an appropriate occupational exposure limit (OEL) for personal measurement is still needed.

Trihalomethanes (THMs) are another group of DBPs that form in swimming pool water in reactions between chlorine and organic compounds. The most common THMs found in the ambient air of swimming pools are chloroform, bromodichloromethane, dibromochloromethane and bromoform.[10] Chloroform and bromodichloromethane are classified by the International Agency for Research on Cancer (IARC) as potentially carcinogenic to humans; chloroform also has a Swedish OEL.

In addition, the warm and humid environment in the swimming pool facilities may also promote the growth of bacteria, which may also cause respiratory health effects.

This thesis describes studies that quantify occupational exposure to trichloramine, THMs and endotoxins in the air of different types of Swedish indoor swimming pool facilities and relates exposure levels to adverse health effects among swimming pool employees, focusing on ocular and respiratory symptoms.

BACKGROUND

Indoor swimming pool facilities

The typical public Swedish swimming pool facility was built in the 1960s or 1970s or sometimes even earlier. Thus, the buildings are getting old and during the next 10 to 20 years, up to 60 new facilities are planned to be built. Conventional Swedish swimming pool facilities usually provide a large rectangular exercise pool, sometimes with an additional smaller training pool and a whirlpool. In recent decades, adventure pools that provide more of an experience to visitors have become more common. Many of the new facilities being planned and built are intended to have several whirlpools, waterslides and wave machines. There is also an intention to provide 'wellness' since most new facilities have an associated gym that offers group training.[11] Many Swedish county councils run indoor swimming pools for habilitation and rehabilitation purposes. Compared to conventional swimming pool facilities, habilitation and rehabilitation facilities are often smaller with fewer visitors. Both habilitation and rehabilitation pools are usually maintained with higher water temperatures for therapeutic purposes. Similarly, adventure facilities also tend to have higher water temperatures, but here it is to engender a feeling of tropical warmth. The American Society of Heating, Refrigerating and Air-Conditioning Engineers (ASHRAE) recommend water temperatures of 24 °C - 29 °C for recreational swimming, while the temperatures of physical therapy pools should be maintained between 29 °C and 35 °C, and from 36 °C to 40 °C for whirlpool and spa pools.[12] The Swedish Association of Local Authorities and Regions (SALAR), recommend water temperatures of 26-30 °C for a regular swimming pool and higher temperatures in training pools, pools for baby swimming, and pools for physical therapy.[13]

General job titles in a public indoor swimming pool facility are lifeguard, swimming school teacher, water aerobics instructor, receptionist, caretaker, cleaner and personnel working in the cafeteria. Regardless of job title, the personnel in Swedish indoor swimming pool facilities often switch between several different work tasks during a work shift, and work tasks can also vary from day to day.

Water quality in indoor swimming pools

Good water quality is required to prevent the spread of infections in indoor swimming pools. The water is therefore filtered and chemically treated to remove particles, disinfect it, and to and maintain a stable pH.

Filtration

Clarity and low turbidity of swimming pool water is maintained by filtration. The numbers of particles and the turbidity in swimming pool water should be kept as low as possible, and in any case the turbidity should not exceed 0.4 Formazine Nephelometric Units (FNU). To improve filtration efficiency, flocculants are often added which cause smaller particles to aggregate into bigger complexes that are more easily filtered out.[9] Filter maintenance is crucial to achieve effective filtration of swimming pool water. Some filters are cleaned by changing the filter media, but most filters need to be regularly backwashed to function properly. Backwash means that a certain amount of the circulating water is pumped backwards through the filter, whereby the filter is cleaned of aggregated material.

Sand filters are the most common filter type used in Swedish swimming pool facilities; they may be of the open or closed type. Closed filters are enclosed in a metal container while the open ones consist of a concrete container filled with a layer of gravel in the bottom and a sand bed on top. Sand filters usually ensure good water quality when they are combined with a flocculent and cleansed using backwash methods.[14]

Disinfection

Disinfection of pool water is necessary to eliminate microorganisms and prevent the spread of infections. Chlorination is the most common method of disinfection used in indoor swimming pools. Chlorine is known to be effective against most microorganisms; it is often added to the pool water as sodium hypochlorite (NaClO) or calcium hypochlorite $Ca(ClO)_2$ but some facilities use chlorine gas (Cl_2) derived from sodium chloride. In the pool water, chlorine exists as hypochlorite ions (ClO^-) and hypochlorous acid (HClO). The "free chlorine" from these substances provides the disinfectant power. To reduce the amount of disinfectant used in swimming pools, a supplementary treatment with ozone or ultraviolet (UV) light can also be used.[5]

Both hypochlorite ions and hypochlorous acid are pH-dependent. The higher the pH, the less HClO is formed, while ClO^- ions have a lower disinfecting ability compared to HClO leading to a lower disinfecting effect with an increased pH value (> 8). To maintain the disinfection effect at a higher pH, more chlorine needs to be added. A higher pH also increases the formation of unwanted disinfection bi-products such as trihalomethanes (THMs).[5] However, the pH of the water should not be too low (< 7) either, as this can promote the formation of chloramines, another group of unwanted disinfection bi-products.[15] The recommended pH range for chlorine-based disinfection in Sweden is between 7.2 and 7.6,[9] while the WHO guidelines indicate a pH range between 7.2 and 7.8.[5] Correction of pH in indoor swimming pools is normally achieved by adding carbon dioxide or hydrochloric acid.[14]

Free chlorine (ClO^- and HClO) is effective at eliminating pathogens in the swimming pool water. However, chlorine also reacts with nitrogen-containing substances such as urea, sweat, ammonia and skin flakes, all of which enter the pool water from bathers. Chloramines (monochloramine, dichloramine and trichloramine) are formed in the reaction between free chlorine and the nitrogen-containing impurities and are referred to as combined chlorine. As the content of combined chlorine increases, the level of free chlorine decreases and the disinfectant effect deteriorates. The levels of combined chlorine are most effectively reduced by adding fresh water. The concentration of organic compounds in swimming pool water is generally low and considered to be a negligible part of the combined chlorine. Swedish guidelines for levels of free and combined chlorine at different pH and temperatures are shown in Table 1.[9]

Table 1. Swedish guidelines for chlorine levels in pool water at different pH and temperatures.[9]

Parameters	Chlorine (mg Cl_2/l)		
	pH 7.2	pH 7.4	pH 7.6
Free chlorine at water temperatures < 35 °C	≥ 0.4	≥ 0.5	≥ 0.6
Free chlorine at water temperatures > 35 °C	≥ 0.8	≥ 0.9	≥ 1.0
Combined chlorine at all water temperatures	≤ 0.3	≤ 0.3	≤ 0.3
Total chlorine at all water temperatures	≤ 2	≤ 2	≤ 2

Occupational exposure in indoor swimming pool facilities

Working in the pool area in an indoor swimming pool facility often means exposure to high humidity and air temperature, as well as noise. Other hazards in indoor swimming pool facilities for both visitors and employees include exposure both to microbial contamination and chemicals. Many of the microorganisms that occur in indoor swimming pool facilities originate from the bathers, often from fecal contamination, while some microorganisms are naturally present in the environment. Chemical exposure can occur from disinfectants, chemicals for pH correction and coagulants used to improve the removal of particles by filtration.[5] Cleaning chemicals, often used daily, are another source of occupational exposure for some of the swimming pool employees. In addition to the direct and intentional use of chemicals in swimming pools, there is also a secondary formation of chemical compounds in the pool water, so-called disinfectant by-products (DBPs).

Disinfection by-products (DBPs)

Chlorination is a common and effective method for disinfecting swimming pool water, but chlorine can also react with other chemicals in the water, giving rise to DBPs. There are many different types of DBPs present in chlorinated pool water. Trihalomethanes are the group of organic by-products most prevalent in chlorinated pool waters; the most common is chloroform ($CHCl_3$). Other organic DBPs are also present such as dichloroacetonitrile (CH_2Cl_2N), cyanogen chloride (CNCl), dichloroacetic acid ($CHCl_2COOH$), trichloroacetic acid (CCl_3COOH), and dichloro-methylamine (CH_3NCl_2).[16-18] Inorganic by-products in the form of chloramines and chlorate have also been detected in swimming pool water.[19]

There are several factors affecting the formation of DBPs in swimming pool water: water temperature and pH; degree of water chlorination; contamination of water by organic and inorganic matter; and nitrogen-containing compounds. The extent of the emission of DBPs from the water to the indoor swimming pool air is determined by the concentration of DBPs in the water and their volatility, the number of bathers and their behaviour, ventilation, water temperature, and water circulation.[2, 4, 20-23]

The populations relevant to epidemiological studies covering health effects and DBPs in swimming pool environments are mainly pool workers, professional swimmers and children. Swimming pool workers and elite swimmers spend a lot of time in swimming pool facilities, and children are particularly vulnerable to environmental insults. Swimmers also tend to have a higher rate of pulmonary ventilation during their workouts, which can lead to a more comprehensive airway exposure for volatile DBPs.

Chloramines

Chloramines are a group of inorganic DBPs formed during the reaction between free chlorine and nitrogenous matter emitted from bathers (urea, sweat, skin squama, cosmetics, etc.).[22, 24]

$ClO^- + H_2O$	$\longleftrightarrow$	$HOCl + OH^-$	Free chlorine
$NH_3 + HClO$	$\longleftrightarrow$	$NH_2Cl + H_2O$	Monochloramine
$NH_2Cl + HClO$	$\longleftrightarrow$	$NHCl_2 + H_2O$	Dichloramine
$NHCl_2 + HClO$	$\longleftrightarrow$	$NCl_3 + H_2O$	Trichloramine

The formation of chloramines is dependent on water pH and the ratio between chlorine and nitrogen (Cl^-/N). Trichloramine (NCl_3) is the dominant form at lower pH values (< 8) and when the ratio between chlorine and nitrogen increases. Different chloramines have different water solubility and volatile properties. Trichloramine is released from pool water about 100 times faster than dichloramine ($NHCl_2$) and about 300 times faster than monochloramine (NH_2Cl), which makes trichloramine the chloramine most commonly found in indoor swimming pool atmospheres.[25] The main route of exposure for chloramine is considered to be inhalation. However, to date, there have been no scientific studies that have examined dermal uptake of chloramines.[26]

Trichloramine has been shown to be a strong irritant and lachrymator.[27] Chloramines are known to interact with the mucous membranes of the eyes and upper airways causing irritation.[28] In-vitro studies have also documented the ability of chloramines to interfere with the junctions between the epithelial cells in the mucous membrane thus increasing permeability.[29] Health effects associated with exposure to trichloramine in indoor swimming pool facilities are mainly acute effects such as irritation of the eye and upper airways and nausea, but asthma has also been related to trichloramine exposure. Previous studies on trichloramine exposure and its health effects in workers at indoor swimming pool facilities have mainly reported ocular and upper airway symptoms at different levels of trichloramine. Workers in an indoor swimming pool facility in Taipei, Taiwan, described symptoms of sore throat and phlegm at an average air trichloramine concentration of 60 $\mu g/m^3$.[30] In a hotel indoor water park resort, lifeguards reported work-related coughs and ocular irritation at a measured mean trichloramine concentration in air of 440 $\mu g/m^3$.[31] In an Italian study, workers exposed to trichloramine levels over 500 $\mu g/m^3$ experienced ocular symptoms, runny noses and loss of voice.[32] Moreover, three studies found average trichloramine concentrations at 110, 560 and 670 $\mu g/m^3$ in indoor swimming pool atmospheres and could associate cumulative trichloramine exposure with ocular and upper airway symptoms among workers in the pool facilities.[1-3] In addition to mucosal symptoms, exposure to trichloramine concentrations of 100-570 $\mu g/m^3$ in swimming pools also seems to be a trigger for pre-existing asthma and an onset for occupational asthma.[4, 33]

In 2006, WHO recommended a reference value for trichloramine of 500 µg/m³.[5] Sweden has not established an occupational exposure limit (OEL) for trichloramine in air, but the Nordic Expert Group for Criteria Documentation of Health Risks from Chemicals (NEG) recommended in 2019 a health-based OEL of 100 µg/m³ for personal measurements, and a corresponding value of 200 µg/m³ for stationary measurements in swimming pool facilities.[34] The Public Health Agency of Sweden (Folkhälsomyndigheten) has established a recommended reference value for stationary sampling of 200 µg/m³ for visitors in indoor swimming pools.[9] In 2011, both Germany and Switzerland adopted 200 mg/m³ as a guide value for trichloramine in air[6, 7] and in 2012, the French agency for food, environmental and occupational health and safety presented a recommended reference value of 300 µg/m³.[8]

Trihalomethanes (THMs)

Trihalomethanes (THMs) are formed when free chlorine reacts with organic materials that occur naturally in the water or originate from bathers.[35] Chloroform ($CHCl_3$) is the most common THM in chlorinated freshwater but bromodichloromethane ($CHCl_2Br$), dibromochloromethane ($CHClBr_2$) and bromoform ($CHBr_3$) also occur and were first discovered by Rook in 1974.[10] The brominated THMs are formed from bromine occurring naturally in the water. Sea-water contains more bromine than freshwater resulting in a higher concentration of brominated THMs in pools using chlorinated sea-water.[36, 37] THMs are generally considered volatile, but chloroform is the most volatile THM found in chlorinated swimming pools.[25]

THMs have been detected in both plasma and alveolar air collected from swimmers and employees at indoor swimming pool facilities.[38-40] Previous studies have also evaluated exposure pathways of THMs in the form of dermal uptake, inhalation, and oral exposure among swimmers. Inhalation accounted for 56-76% of total THM uptake among swimmers and was thus considered to be the main route of exposure.[20, 41-44] Inhalation also seems to be the most significant route of exposure for THMs associated with an increased risk of cancer.[45]

The International Agency for Research on Cancer (IARC) has classified chloroform and bromodichloromethane as possibly carcinogenic to humans (group 2B), while dibromochloromethane and bromoform are currently not considered carcinogenic to humans (group 3). Previous studies using multi-pathway health risk assessment models have found an increased life-time cancer risk for swimmers from THM concentrations in water.[46-49] A significant increased risk of developing bladder cancer has been seen among swimmers compared to non-swimmers.[50] Both elite swimmers and coaches exposed to THM concentrations of 59-397 µg/m^3 in the air above the water surface and of 28-390 µg/m^3 in the air surrounding the pool area were found to have an elevated cancer risk.[51] However, one should bear in mind that calculated cancer risks are often based on many assumptions.

For chloroform in air, Sweden has a standard 8-hour OEL of 10 000 µg/m^3 defined by the Swedish Work Environment Authority (SWEA).[52] The Public Health Agency of Sweden (Folkhälsomyndigheten) also has a recommended limit value of 100 µg/L for THMs in chlorinated swimming pool water.[9]

Endotoxins

Endotoxins are components of the outer cell membrane of gram-negative bacteria. They are emitted from the cell membrane when bacteria replicate and when they die. Endotoxins are found in most forms of organic dust mainly originating from animal feces and contaminated plant materials. Hence, a large part of the occupational exposure to endotoxins takes place in the agricultural sector. Inhalation is the main pathway of occupational exposure since endotoxins become airborne when organic materials are manufactured or handled.[53]

The most common endotoxins are lipopolysaccharides (LPS) that consist of molecules containing lipids and polysaccharides. The toxic properties of LPS are mainly due to the lipid moiety, a phosphoglycolipid called “lipid A”. The overall structure of the LPS can vary between different species of gram-negative bacteria but “lipid A” has more or less the same structure regardless of bacterium.[53] Endotoxins are known to be heat stable where inactivation of LPS occurs first at a temperature of 177 °C.[54]

Previous studies on occupational exposure to endotoxins cover the animal production industries; e.g. those involving cattle[55], poultry[56, 57], swine, and wool[57]; but also plant production; e.g. cotton[56-58], grain[56, 57, 59] and wood.[56, 57, 60-63] In a hotel indoor water-park resort, lifeguards reported work-related coughs and ocular irritation at endotoxin concentrations of 18 to 84 endotoxin units (EU)/m^3 with the highest air concentrations near the wave pool (84 EU/m^3) and the leisure river (67 EU/m^3).[31] A case study reported two outbreaks of granulomatous lung disease among lifeguards in an indoor swimming pool facility exposed to mean concentrations of endotoxins in air of 28 and 76 EU/m^3.[64]

Occupational exposure to increasing levels of endotoxins via inhalation can cause acute symptoms such as cough[65], as well as nasal and asthma symptoms.[66] Toxic pneumonitis is a fever reaction with influenza-like symptoms that may occur a few hours after heavy exposure to endotoxins.[67, 68] If the endotoxin exposure is chronic it may lead to chronic bronchitis and reduced lung function. Affected spirometric values in the form of decreased forced expiratory volume in 1 second (FEV1) are often found among subjects exposed to airborne endotoxins.[69] Acute effects of endotoxin concentrations over 90 EU/m^3 have been seen as a decrease in FEV1 over a 6 h work shift in an experimental study including otherwise healthy volunteers exposed to cotton dust with endotoxin concentrations of 60-7790 EU/m^3.[70] Long-term effects of endotoxin exposure causing an annual decrease in FEV1 were reported among workers in the animal feed industry exposed to endotoxin concentrations of 2-4700 EU/m^3.[71, 72] From these studies, 90 EU/m^3 was set as a recommended occupational exposure limit value for long-term air endotoxin exposure.[53, 69] In later years, long-term mean endotoxin exposures below 100 EU/m^3 have been associated with adverse respiratory health effects.[73]

Occupational hygiene

Occupational hygiene is the practice of identifying hazardous chemical, physical and biological agents in a workplace. Identified hazards that can cause disease or discomfort should be, evaluated by occupational hygienists to determine the extent of any risk due to exposure to such hazardous agents. Controlling and preventing these risks is also of the utmost importance to prevent chronic or acute ill-health.[74]

Occupational exposure

The term "exposure" in the field of occupational hygiene refers to the level of contact with a hazardous agent and its entry into the body via the lungs, gastrointestinal tract or through the skin.[75] Occupational exposure has traditionally focused on airborne exposure, but dermal exposure and ingestion are other pathways that should also be considered.[76] Exposure is normally quantified as the level of contaminant in contact with the body, often measured in mg/m^3 of air in the breathing zone. It can also be quantified as the level of contaminant or metabolite inside the body in µg/L of blood or urine.[75]

Measurement of occupational exposure

Occupational exposure in the form of chemicals in workplace air is often measured using personal and/or stationary sampling. Personal exposure levels are preferably measured by attaching a sampler to the worker's clothing within their breathing zone, while stationary sampling is carried out with measuring devices placed at fixed positions.[76] There are different kinds of samplers available to collect chemicals in workplace air, e.g. a filter placed in a monitor or a sorption tube.[77] The sampler can be connected to a pump where a controlled amount of air is drawn through the sampler but the development of passive samplers also allows monitoring exposure to gaseous species without the need of a pump. The gaseous or particulate matter trapped in the sampler is then analyzed and quantified in a laboratory.[75]

The results from exposure measurements are often used for comparison with occupational exposure limits and can be helpful in evaluating improvements to the work environment. The results can also be used in general monitoring programs, surveillance and comparisons of different sampling or analytical methods.[77]

Occupational exposure limits (OELs)

In 1969, the Swedish Institute of Occupational Medicine published their first list comprising about 70 occupational exposure limits (OELs).[78] During the 1970s the awareness of chemical risks in the work environment increased markedly. The Swedish list of OELs, published by the Swedish Work Environment Authority (SWEA), still exists today and is regularly updated. Over the years, several limit values have been added, existing values have been reduced, and some have been removed.[79] The latest version, published in 2018, contains about 500 chemical substances with their associated OELs.[52]

The earliest Swedish OELs were influenced by the American Conference of Governmental Industrial Hygienists' (ACGIH's) list of TLVs, but in 1978 Sweden developed its own process to establish national OELs. Sweden has also included so-called pragmatic or administrative OELs, which are based not only on knowledge about detrimental health effects of the substance, but also on technical and/or socioeconomic factors. The OELs are set by SWEA in a process that takes place in four main steps: prioritization of substances, collection of data, impact assessment and decisions. The OEL is often based on scientific information from criteria documents found in Work and Health produced by the Nordic Expert Group for Criteria Documentation of Health Risks from Chemicals (NEG).[80]

The Swedish OELs are defined as the highest acceptable concentration for chemicals in workplace air and are in most cases binding by law. For some substances, however, there are indicative short-term limits. Swedish OELs apply to dust, smoke, fog, gas and steam and are defined as either an 8-hour OEL or a short-term limit, or as a recommended short-term limit. For comparison with an 8-hour OEL, the sampling time should preferably last during a whole working day or at least 75% of the working day. The short-term limit relates to an exposure lasting 5 to 15 minutes. The chemical of interest should preferably be measured at a maximum of 0.3 meters from the workers nose and mouth, i.e., within his or her breathing zone.[52]

Occupational medicine

Occupational medicine is the science of how chemical, physical, biological and physiological factors in the working environment affect human health, and what preventive measures should be taken to reduce health risks over the course of a working life. The discovery and treatment of what are now commonly known as classic occupational diseases, such as lead poisoning, mercury poisoning and silicosis, have formed the basis and development of occupational medicine. During the 20th century, there has been a continuous improvement in the industrial environment and in the health and safety of the working population; this has been partly due to a shift from manual to automated work. In later years, the focus in occupational medicine has changed from acute injuries to chronic conditions such as airway diseases.[81]

The ocular and respiratory tract

The ocular and respiratory tracts are prime targets for airborne chemicals, both in the form of vapours and as liquids in aerosols. There are several different factors that affect a chemical's ability to enter and affect the body. A chemical's volatility, structure, molecular size, as well as its water and fat solubility, and its reactivity determine the sensitivity of the target site and the chemical's ability to interfere with water based bio-fluids or its transportability across lipid membranes. Chemicals can react with the ocular surface and enter the respiratory tract where they may stimulate chemical receptors, trigger immunological defence mechanisms or cause structural and biochemical changes.[82]

Eye irritation

The outer eye has an ocular surface comprising the cornea and conjunctiva that is made up of epithelial cells covered by a tear film. The tear film is produced by different glands and consists of three layers; an inner mucus layer, an aqueous-mucin layer, and an outer oily lipid layer. The tear film provides the cornea with a smooth surface and prevents water evaporation preserving the moist surface of the outer eye.[83]

The destabilisation and break-up of the tear film are known to cause eye irritation. Eye irritation is a general term that refers to local effects in the mucosa of the eye or surrounding tissues such as the eyelid. While some symptoms can be severe, such as burning, irritation, smarting, red eyes, pain or secretory phenomena, others are less severe, such as dryness, discomfort or itchiness. There can also be many different causes of eye irritation such as responses to allergens, infections, diseases, dry eyes, or digital eye strain, and other irritants.[82, 84] Symptoms of eye irritation may arise in conjunction with chemical sensory irritation when airborne chemicals trigger chemoreceptors of the eye, which then activate the nervous system. A chemical trigger of the nervous system may also induce reflexes such as blinking.[85] Several water-soluble chemicals, for example aldehydes, have been seen to interact with the surface of the eye and cause eye irritation.[86, 87]

Symptoms of eye irritation are often self-reported as subjective complaints but there are more objective methods available. A slit lamp can be used to examine the appearance and the stability of the tear film and assess any damage to the conjunctival epithelium.[88]

Rhinitis and upper airway irritation

The upper respiratory tract (URT) consists of the nose, pharynx and larynx. The URT works as a conditioner and as the body's first line of defence. As air passes through the URT it becomes humidified and heated to body temperature; the nose also filters particles larger than 10 µm from the passing air. The respiratory epithelium is made up of epithelial cells and secretory cells. The secretory cells produce the mucous membrane containing inflammatory mediators, interferons, and immunoglobulins.[89]

Rhinitis is an inflammation of the mucous membrane of the nose with intermittent or persistent symptoms such as sneezing, itching, runny nose and nasal congestion, sometimes in combination with variable nasal airflow limitation and/or hypersecretion.[90] Rhinitis can be caused by an infection, by irritants or allergens.[91]

The upper airways are, just like the ocular surface, a target for chemical sensory irritation. Chemicals can trigger chemoreceptors of the upper airways leading to a nervous system activation causing immunological defence mechanisms, inflammatory symptoms similar to those that occur with rhinitis, and also induced reflexes such as sneezing and coughing.[82, 85]

Asthma

The lower respiratory tract (LRT) is situated below the trachea and is divided into conducting airways (trachea, bronchi and non-respiratory bronchioles) and the respiratory unit (respiratory bronchioles and alveoli). Just like the URT, the bronchi and bronchioles in the LRT consist of the respiratory epithelium with epithelial and secretory cells producing the protective mucous membrane containing cells from the immune system.[89] The alveoli are protected by a liquid layer called the lung epithelial lining fluid (ELF) containing fatty acids, proteins and phospholipids.[82]

Asthma is an obstructive pulmonary disease characterized by a chronic inflammation triggered by irritants and allergens (bronchial hyper-reactivity) and is characterized by reactions in the conducting airways. An immunological response, mainly from lymphocytes and eosinophils, leads to an increased production of mucus and a reversible constriction of the smooth muscles of the airways (bronchospasm) resulting in an obstructed airflow through the airways.[89] Typical symptoms of asthma are shortness of breath, wheezing, tightness in the chest and cough.[92]

Occupational asthma and rhinitis

Asthma is a chronic disease with approximately 260 million sufferers worldwide.[93] Approximately 15% of all asthma occurring in adults is considered to be work-related.[94] Earlier studies among workers exposed to sensitizing agents have indicated the incidence of work-related rhinitis to be two to four times more common than work-related asthma. The majority of patients diagnosed with work-related asthma also suffered from work-related rhinitis[90] and a history of work-related rhinitis has been pinpointed as a risk factor for the development of work-related asthma.[95-98] Work-related asthma and rhinitis can be divided into occupational asthma/rhinitis (OA/OR) and work-exacerbated asthma/rhinitis (WEA/WER).[90, 99] OA and OR is caused by exposure to sensitizers or irritants in the workplace and can be allergic or non-allergic.[90, 100, 101]

The allergic form of OA and OR can be Immunoglobulin E (IgE)-mediated caused by a wide variety of high molecular weight (HMW) (≥ 10 kDa) agents or triggered by low molecular weight (LMW) (≤ 10 kDa) agents with an IgE-mediated mechanism.[90, 99] HMW agents include glycoproteins originating from plants or animals and can provide occupational exposure as, for example, flour among bakers[102] or latex among healthcare workers.[103] LMW compounds are usually small and need to act as haptens conjugating with host proteins in order to elicit an IgE sensitization response[104-106], but the mechanism is not fully understood.[107] There are many LMW agents associated with an IgE mediated antibody response: platinum salts used in platinum refineries[108]; salts containing rhodium[109], nickel, chrome and cobalt[110]; acid anhydrides as hardeners in epoxi resins[111]; reactive dyes in the textile industry[112]; persulphates among hairdressers[113]; and wood dust among carpenters and furniture makers.[114] One of the most common LMW agents causing OA are diisocyanates used as hardeners in polyurethane foam and as hardeners in spray paint and adhesives.[104] Specific IgE antibodies can be detected in up to 55% of workers with confirmed OA exposed to diisocyanates.[104, 115] Allergic OA and OR is preceded by a latency period of months to years after initial exposure.[116, 117]

The non-allergic forms of OA and OR are triggered by the work environment through non-immunological irritant mechanisms and have no latency period. OA and OR can occur with one high level exposure to a respiratory tract irritant such as chlorine gas and cause a so-called reactive airway dysfunction syndrome (RADS) or a reactive upper airway dysfunction syndrome (RUDS). Irritant induced asthma is another form of non-allergic OA and OR with an unknown pathophysiological mechanism, caused by multiple exposures.[90]

Work-exacerbated asthma and rhinitis (WEA/WER) is defined as a pre-existing allergic or non-allergic asthma or rhinitis that is not caused by, but is worsened by workplace exposure.[100, 101, 118, 119] Many various agents can trigger an already existing asthma or rhinitis including irritants such as chemicals, dusts and fumes, and physical factors such as temperature changes as well as second-hand smoke and strong smells.[90]

AIMS

The overall aim of this thesis was to gain an understanding of exposure levels of trichloramine, trihalomethanes and endotoxins in the ambient air of Swedish indoor swimming pool facilities and investigate adverse ocular and respiratory health effects among indoor swimming pool workers. The specific aims are listed below.

- Investigate occupational exposure to trichloramine, THMs and endotoxins in the air of different types of Swedish indoor swimming pool facilities.

- Investigate adverse health effects, focusing on eyes and respiratory effects, among swimming pool employees in Swedish indoor swimming pool facilities.

- Investigate a possible relationship between measured occupational exposures and registered adverse health effects among swimming pool employees to predict safer exposure levels.

MATERIAL AND METHODS

Study design

Epidemiological studies are quantitative and describe the incidence of disease in a population. There are different types of epidemiological study designs. Many epidemiological studies have a longitudinal perspective: a research design that involves repeated observations over short or long periods of time. A cross-sectional study design, on the other hand, has multiple observations originating at one point in time. In longitudinal studies, the exposure is expected to precede the outcome, while cross-sectional studies are snapshots capturing disease and exposure variables at a specific point in time. Cross-sectional designs tend to be more efficient with respect to time and costs, but data from these studies cannot be used to measure disease incidence, since risk or rate calculations require data across a time period. However, cross-sectional data can be used to assess disease prevalence.[120] Sometimes no relationship is found between exposure and disease prevalence. In these cases, an unexposed (control) group can be used to provide information about the expected disease prevalence in the exposed group.[121] The controls should not be exposed to the hazard being studied, but otherwise as similar as possible to that experienced by the exposed group.

In the studies reported here, a cross-sectional design has been used as information regarding exposure and medical status refers to the same point in time. However, repeated observations recording medical data have been made during the single day of the investigation. In addition, retrospective data regarding previously experienced symptoms have also been collected via questionnaires. Unexposed controls (referents) have also been used for comparison.

Study objects

A total of 23 Swedish indoor swimming pool facilities of three different types were included in the studies comprising this thesis. During the period 2007-2009, data were collected at eight conventional swimming pool facilities. During the same period, referents comprising 50 office workers unexposed to swimming pool air were recruited from municipal offices in the same cities whereas the conventional swimming pool facilities were located. During the years 2012-2014, ten habilitation and rehabilitation swimming pool facilities participated; during 2019-2020, further data were collected at five adventure swimming pool facilities. The characteristics of the different facilities included in the study are presented in Table 2.

Table 2. Number of employees, annual visitors and pool information for the different types of indoor swimming pool facilities included in the studies (Papers I-IV).

Type of swimming pool facility	Employees (n)	Annual visitors (n)	Swimming pools (n)	Pool types	Water temp. (°C)	Water treatment
Conventional (n = 8)	4-35	18 000-300 000	1-3	Exercise pool Training pool Whirlpool Children's pool	27-37	Chlorine Sand filter UV
Habilitation and rehabilitation (n = 10)	-	4000-50 000	1-2	Rehabilitation pool	34	Chlorine Sand filter UV Ozon
Adventure (n = 5)	10-32	140 000-500 000	5-7	Exercise pool Training pool Adventure pool Whirlpool	27-38	Chlorine Sand filter Carbon filter UV

The conventional swimming pool facilities had 4 to 35 employees and an annual number of visitors of 18 000-300 000. They had from one to three pools: each had an exercise pool and some also had a training pool and/or a whirlpool and a specific pool for children. The water temperatures ranged between 27-37 °C. Example of a Swedish conventional swimming pool facility is shown in Figure 1.

Figure 1. Example of a Swedish conventional swimming pool facility.

Information on the number of employees at the habilitation and rehabilitation facilities are missing, but they had approximately 4000-50 000 visitors a year. They offered at least one rehabilitation pool, but in addition, some of the facilities also had a smaller pool. They all held a water temperature of 34 °C. Example of a Swedish habilitation and rehabilitation swimming pool facility is shown in Figure 2.

Figure 2. Example of a Swedish habilitation and rehabilitation swimming pool facility.

The number of employees in the adventure facilities ranged between 10-32 and they received approximately 140 000-500 000 visitors a year. The adventure facilities had 5-7 pools: for exercise and training, whirlpools and adventure pools with, for example, waterfalls, waterslides, rapids and wave pools. The water temperatures in the adventure facilities were held between 27-38 °C. Example of a Swedish adventure swimming pool facility is shown in Figure 3.

Figure 3. Example of a Swedish adventure swimming pool facility.

All swimming pools included in the study were primarily disinfected using chlorine and were also equipped with sand filters. As a complement, UV-treatment was used in one or more of the pools in two conventional facilities, seven habilitation and rehabilitation facilities, and all adventure facilities; ozone was used in one of the habilitation and rehabilitation facilities. The ventilation systems in all facilities were operated with heat recovery.

Among the 23 facilities, a total of 117 workers were included in the studies. To be included, the workers had to work at least 4 hours (equivalent to 50% of a full work shift) and be unexposed to swimming pool air 48 hours before taking part in the investigations. The participants worked during a day or evening shift.

Ethical considerations

The Helsinki Declaration is a central set of research ethics guidelines that was adopted by The World Medical Association (WMA) in 1964. The declaration concerns the ethical principles involved when people are subjects of research. The declaration is not legally binding but has a significant influence on national legislation. One of the basic principles is that the individual's rights and needs are more important than the interests of science and the society. The declaration also emphasizes the importance of an individual's consent being given before initiating data collection and analysis and possible storage of samples. The individual should also be treated with respect, and all data regarding the individual should be treated as anonymous and confidential.[122]

According the studies included in this thesis, the study participants were given written and verbal information before data collection was initiated. The information gave the participants knowledge of the purpose of the study, that their participation was voluntary and that they had the right to end their participation at any time. They were also informed about their right to know which of their data were used in the study. Before data collection was initiated, they also signed an agreement to participate. A physician assessed the results of medical examinations, and any abnormal results were followed up if necessary and in some cases the participant was referred to specialist care.

The Helsinki Declaration is incorporated in the Ethical Review Act - a Swedish law regulating the ethics of research that involves humans. The law requires an application for ethical review, which applies to all research involving living persons, the deceased, human biological material, and research handling sensitive data about people.[123] The studies included in this thesis were approved by the Regional Ethical Review Board in Uppsala, Sweden (Papers I and II: decision no. 2006/349, Paper III: decision no. 2012/170, Paper IV: decision no. 2018/314).

Exposure assessment and chemical analysis

Personal and stationary air sampling

The swimming pool employee's individual levels of exposure were documented via personal sampling in the breathing zone by placing a trichloramine filter cassette and a THM sorbent tube on their shirt collar (see Figure 4). To be able to follow their working day and document their different work tasks and duration, the participants were also asked to keep a work diary during the time when measurements were taken. The work diaries were then used to categorize the swimming pool workers in the conventional facilities and the habilitation and rehabilitation facilities into different exposure groups. A low-exposure group was defined as those employees that spent no time in the pool area; a medium exposure group - those with a working day less than 50% in the pool area; and a high exposure group - those who spent 50% or more of the day near the swimming pools.

Figure 4. Personal air sampling of trichloramine and trihalomethanes.

Stationary 8-hour sampling was executed to document the ambient levels of trichloramine, THMs and endotoxins in the air and their variation among the facilities. Trichloramine filter cassettes, THM sorbent tubes and endotoxin filter cassettes were placed at different fixed positions around the facilities, 0-1 m from the pool side at height of approximately 1.5 m (see Figure 5). Personal and stationary samples of trichloramine and THMs were taken in all facilities, while only stationary endotoxin samples were taken in the adventure facilities. In conventional and in habilitation and rehabilitation facilities, stationary sampling was also performed in the reception areas. In addition to the 8-hour sampling, stationary sampling in the conventional facilities was also executed during specific activities, such as swimming school and baby swimming.

Figure 5. Stationary air sampling of trichloramine and trihalomethanes.

Trichloramine – sampling and analysis

In 1995 Hery et al. presented a sampling and analytical method suitable for trichloramine in the ambient air of indoor swimming pools.[22] The sampler used to collect trichloramine in air consisted of a filter cassette containing two impregnated quartz fiber filters (37 mm): the first a sampling filter; the second, a backup filter. The filters were impregnated with a solution of sodium carbonate and diarsenic trioxide. Sodium carbonate was used to obtain a high pH to decompose the chloramines collected on the filter into ammonia and hypochlorite. Diarsenic trioxide then reduced the hypochlorite into chloride (Cl^-). A Teflon pre-filter was placed in front of the sampling filter to prevent contamination from airborne water droplets. After sampling but prior to analysis, the chlorides were extracted from the filters using double distilled water. The chloride content of the filters was then analyzed with compressed ion chromatography and conductivity detection adapted for anions.[22]

Ion chromatography (IC) is a well-known type of liquid chromatography (LC) used to analyze anions (negatively charged ions) or cations (positively charged ions) of organic or inorganic origin in various types of sample, including environmental samples. The technique separates ions of interest from samples in an ion exchange based on their molecule charge.[124] The samples need to be extracted as the analyte must consist of solute ions. The extracted sample is then injected into a carrier fluid (the mobile phase) and passed through a fixed stationary material (the stationary phase). The mobile phase (eluent) consists of a solution often comprising hydroxide or a carbonate and the stationary phase consists of an anion or cation exchange resin, accommodated in a column. As the mobile phase moves through the stationary phase, the sample ions are more or less attracted to the oppositely charged ions in the stationary phase. Those more attracted to the stationary phase move more slowly through the column, while those less attracted move faster. Thus, the sample ions move through the column at different speeds and so separate from one another. In addition to electric charge, the separation is also affected by the composition of the eluent, pH, temperature and flow rate. Before the ions separated in the mobile phase reach the detector, they pass through a suppressor. Ion suppression is used to suppress the conductivity of the mobile phase and enhance the conductivity of the sample ions making them easier to detect. The suppressor consists of an ion-exchange membrane where the eluent is neutralized. The ions are then detected according to their electrical conductivity in a conductivity detector, which consists of a multi-electrode flow cell. In the detector output, each group of ions from the sample separated in the column is presented as a peak at a specific retention time in a chromatogram. The area under each peak is proportional to the mass of the compound passing through the detector.[125]

Sampling of trichloramine in air and its subsequent analysis in these studies was also based on the method published by Hery et al.[22] However, prior to trichloramine sampling some modifications were made to the sampler and rate of airflow during sampling. Hery's filter cassette used for trichloramine sampling contained a Teflon filter in front of the sampling filter to prevent water contamination. The filter cassettes used for trichloramine sampling in the present study did not contain a Teflon filter as the risk of water contamination in this setup was considered to be minimal. To enable sampling during a whole 8-hour working day, and not risk breakthrough, the airflow was decreased from 1 l/min, as used by Hery et al., to 0.25 l/min. A consistent airflow of 0.25 l/min through the trichloramine sampler was maintained using an airflow pump (GSA SG4000, Germany or SKC AirChek 5000, USA) connected to a twin-port sampler (MSA Gemini®, USA). The samples were analyzed in an IC (Triatlon 900 autosampler, Spark, Netherlands) with a conductivity detector (Costech Microanalytical Ltd, Tallin, Estonia). The limit of detection (LOD) of the method was set to 0.213 µg/sample.

Trihalomethanes – sampling and analysis

Sampling airborne volatile organic compounds (VOC) involves pulling air through a sorbent tube followed by an analytical procedure including thermal desorption, gas chromatography (GC) and mass-spectrometry (MS). A number of sorbent tubes with different packing material are available on the market. Tenax and Molecular Sieve are two common packing materials used to sample VOC, but multisorbent tubes with more than one packing material are also available. These multibed tubes contain sorbents with different sorbent strengths and are able to collect a wider range of VOCs.[126]

To identify and quantify the VOCs sampled with the sorbent tubes, automatic thermal desorption (ATD) and GC-MS was used. The sampled sorbent tubes are purged with dry, inert gas before analysis to remove water vapor and air that can interfere with the analysis. The samples are then placed in an ATD apparatus where a two-stage thermal desorption takes place. In the first stage, the VOCs are released from the sorbent tubes as they are heated in a flow of inert gas. In the second stage, the VOCs are cryo-focused onto a cold trap to concentrate them before they are injected into the GC column via a heated transfer line. Temperature, time and gas flow are all factors that affect the thermal desorption and can be adjusted.[126] The GC column is placed in an oven running at a controlled temperature. The VOCs are transported through the column using a carrier gas as the mobile phase. The inner wall of the GC column is covered by the stationary phase with which the VOCs react differently depending on their individual chemical properties. Their different abilities to interfere with the stationary phase separates them from each other leading to different retention times. At the end of the column, the separated VOCs reach the MS where they are ionized and accelerated in an electric field leading to separation by their mass-to-charge ratio. The different ions are then detected by an electric multiplier, which displays the signals as a mass spectrum. The different compounds are identified by comparing the mass spectrum against a library of known masses and their characteristic fragmentation pattern. The different VOCs can be quantified because the area under each peak in the mass spectrum is proportional to the mass of compound passing through the detector.[125]

In the conventional facilities (Paper I) and habilitation and rehabilitation facilities (Paper III), air sampling and analysis of VOCs in the form of trihalomethanes (chloroform, bromodichloromethane, dibromochloro-methane and bromoform) were based on the EPA method TO-17.[126] Air sampling was conducted with Carbotrap™ 300 (Perkin Elmer, USA), a multibed sorbent tube containing three layers of different carbon adsorbents (Carbopack B, Carbopack C and Carbosieve SIII).[127] Carbosieve SIII is a carbon molecular sieve with properties considered to be suitable for sampling halogenated hydrocarbons.[126] In the adventure facilities (Paper IV), the multibed sorbent tube Universal Mix (Markes International, USA) with three different adsorbents (Tenax TA, Carbograph 1TD and Carboxen 1003) was used. The sorbent tube was connected to an airflow pump (GSA SG4000, Germany or SKC AirChek 5000, USA) and a twin-port sampler (MSA Gemini®, USA) operating at an airflow rate of 0.1 l/min. To avoid breakthrough, the maximum sampling time was set to 4 hours. To be able to sample for an entire 8-hour workday, the sampler was changed after 4 hours. The samples were analyzed using an ATD-GC-MS (ATD TM, Perkin Elmer, USA and GC 6890, Hewlett Packard, USA). The limit of quantification (LOQ) was a signal to noise ratio (S/N) larger than 10, except for bromodichloromethane which had a S/N of 4.7. The lowest concentrations for each trihalomethane were well above the LOQ, 0.75 ng per sample for chloroform, and 0.10 ng per sample for bromodichloromethane, dibromochloromethane and bromoform.

Endotoxins – sampling and analysis

Airborne endotoxins are usually measured using a filter mounted in a closed-face, three-piece cassette connected to an air-sampling pump. A number of different filters are used for endotoxin sampling. Binder-free glass fibre filters are recommended as a Swedish standard for quantitative measurement of endotoxins in workplace air[128] and are the filters most often used; but filters consisting of polycarbonate, polyvinylchloride (PVC), polytetrafluorethylene (PTFE) (Teflon®), or cellulose are also available.[129] The type of filter used for sampling can affect the results as materials such as polycarbonate, PVC and PTFE can absorb endotoxins with the risk of giving false low values.[128] To analyze the sampled endotoxins a Limulus Amebocyte Lysate (LAL)-assay is used.[69, 128, 129] Before the LAL-assay can be performed, the sampled endotoxins need to be extracted from the filters, preferably in endotoxin-free water. The LAL-assay technique is based on natural properties of blood cells (amebocytes) found in the horseshoe crab, *Limulus polyphemus*. In contact with endotoxins, the cells react to form a gel. In the LAL-assay a limulus amebocyte lysate is used, a solution containing the limulus amebocytes. There are two types of LAL-assays available for detection and quantification of endotoxins: a turbimetric assay and a chromogenic assay. In the turbimetric assay, the extracted sample is mixed with LAL and in presence of endotoxins the gel formation results in a turbidity that is considered to be quantitatively relative to the concentration of endotoxins. In the chromogenic assay, the extracted sample is mixed with the LAL and a synthetic colourless substrate. If the sample contains endotoxins, the enzyme activating the gel formation also catalyses the substrate resulting in a yellow colour. The colour can be measured via photometry, the resultant measure being a function of the concentrations of endotoxins in the sample.[128]

In Paper IV, the endotoxin sampling was executed on polycarbonate filters (25 mm) placed in a cassette and connected to an airflow pump (SKC AirChek 5000, USA) keeping a constant airflow of 1.5 l/min during an 8-hour work day. The sampled filters were analysed in a LAL-assay with a LOD of 0.005 EU/ml.

Adverese health effects assessment

Questionnaire

Questionnaires are an important instrument for collecting data in epidemiological studies, especially from a large population at a low cost. Information concerning an individual's exposure is often collected using questionnaires. Compared to collecting data via interviews, the responses via questionnaires are often more candid and not affected by the interviewer.[130] The advantages of this method are many but there are some limitations that can introduce bias. The design of the questionnaire is of great importance: questions must be clear in order to avoid underreporting.[120, 131] Answers to questions concerning past exposures can be hard to recall and so introduce a recall bias.[132] Recall bias may also be an issue where those with disease are more likely to report their exposures compared to those without a disease.[120] On the other hand, questionnaires can also allow confounding factors to be controlled by including questions covering background factors, such as smoking.

In Papers II, III and IV, a questionnaire was used to collect medical data parameters covering ocular, and upper and lower airway symptoms experienced the previous seven days, the previous three months, and the previous year. The participants were also asked to report information regarding sex, age, smoking habits, time in present job, asthma at any time, and allergic symptoms during childhood. The questionnaire used in this work is a combination of different questionnaires used in earlier occupational exposure studies.[133-135]

Fraction of exhaled nitric oxide (FeNO)

Nitric monoxide (NO) is found in many human organs and is naturally produced in the lung.[136] Thus, NO is also present in exhaled breath were it can be used as a clinical biomarker. Deviating concentrations of NO can be a part of the pathophysiology of lung diseases, including asthma.[137] The NO concentration in exhaled breath is called the 'fraction of exhaled nitric monoxide' (FE_{NO}) and is measured by a single 10-second blowout into a FE_{NO} instrument. Elevated NO levels in exhaled breath can be caused by eosinophilic airway inflammation, a condition that often occurs in asthma. The American Thoracic Society has suggested that FE_{NO} values greater than 50 ppb may be an indication of eosinophilic asthma.[123] Increased FE_{NO} levels have been reported in association with occupational exposure to high molecular weight agents such as endotoxins or isocyanates - agents known to cause a neutrophilic airway response or eosinophilic airway inflammation.[138]

The main causes of increased FE_{NO} levels are atopy, airway inflammation, and asthma, but eating nitrate-rich foods before a FE_{NO} measurement may also increase the concentration. On the other hand, factors like spirometry, smoking, exercise, and bronchoconstriction may decrease FE_{NO} concentrations.[123]

In Papers II, III and IV; a NIOX Mino instrument (Aerocrine AB, Solna, Sweden) was used to measure FE_{NO} levels in exhaled breath. The measurements were made with a flow rate of 50 ml/s and the results were expressed in parts per billion (ppb). The FE_{NO} measurements were performed before and after a work shift, immediately before the spirometry test. Measurement of FE_{NO} levels in exhaled breath is shown in Figure 6. Factors known to affect FE_{NO} levels, such as a recent intake of nitrate-rich foods, smoking habits, and exercise, were documented.

Figure 6. Measurement of fraction of exhaled nitric oxide (FeNO) levels in exhaled air.

Spirometry

Spirometry is a breathing test measuring lung function. It measures how much air you can exhale after full inspiration and how easily and fast the expiration is performed. Spirometry is a screening test used as part of the diagnosis of various respiratory diseases, for example, asthma and chronic obstructive pulmonary disease (COPD). The parameters being measured in spirometry are vital capacity (VC), forced vital capacity (FVC), and forced expiratory volume in one second (FEV1). VC is the maximum volume of air exhaled in one slow expiration starting from full inspiration. FVC is also the maximum volume of air in an expiration after full inspiration, but this expiration needs to be forced. FEV1 is the volume of air exhaled during the first second when FEV is measured.[139] The ratio between FEV1 and FVC can also be calculated and represents the percentage of the maximum lung volume that can be expired in one second. The results from VC, FVC and FEV1 are expressed in liters and presented as a percentage related to normal values based on ethnicity, age, sex and body length.[140] The normal values used in Sweden are the European Coal and Steel Community (ECSC) reference values compiled in 1993 and recommended for European countries by the European Respiratory Society (ERS)[141], and values originating from two older Swedish sources.[142, 143] In 2012, an ERS task force presented new global spirometry reference values including those for several different ethnicities and a larger age span compared to the older reference values.[144] Traditionally, 80% of predicted VC, FVC and FEV1 values have been used as a lower limit of normal (LLN); a ratio of FEV1/FVC below 70% has been defined as airflow obstruction. Thus, the use of fixed thresholds has been questioned since the lower limit has been shown to vary with age.[145] A more accurate way of setting a LLN is, according to the ATS and ERS, to use the fifth percentile.[146] Spirometry also provides a flow-volume curve, which, in addition to individual values, should be assessed and included in the evaluation of the results.

In studies II, III and IV; a hand-held PC-based spirometry flow sensor (Spirare, Diagnostica, Oslo, Norway) was used to test lung function in accordance with the American Thoracic Society guidelines.[139] Measurement of lung function using spirometry is shown in Figure 7. The results were presented as a percentage of normal values for FVC and FEV1 based on sex, age, and body length given in a Swedish reference.[142, 143]

Figure 7. Measurement of lung function using spirometry.

Peak expiratory flow (PEF)

Peak expiratory flow (PEF) is the highest flow measured in a maximum expiration starting from full inhalation. PEF can be read from the flow-volume curve obtained in a spirometry test but it can also be measured via portable monitoring instruments. The PEF results from a spirometry test are expressed in L/s while PEF values from portable monitoring use L/min as the unit.[139]

In the diagnosis of occupational asthma, it can be valuable to measure airflow limitation in relation to occupational exposure. This is normally done as an evaluation of FEV1 in a provocation test, but changes in the airways at a workplace setting are easiest to assess from serial PEF measurements using a portable instrument.[147] It is recommended to take serial PEF measurements at least four times a day[148] with three serial measurements each time.[139] PEF levels have been shown to have a normal variation during the day where morning values are lower than those measured later during daytime.[149, 150] Normal diurnal PEF variations of 6%

have been reported.[151] Compared to controls, a greater diurnal PEF variability has been seen in individuals with COPD and asthma, as well as in smokers.[149] A PEF variability of more than 20% during the day has been used as an indication both of general asthma[152], and occupational asthma.[147, 153] Mean values of collected serial PEF measurements during a work day can also be compared to an average value measured during a day away from work.[154]

In study III, PEF measurements were made on each swimming pool worker using a Mini Wright Peak Flow Meter (Airmed, London, UK). Three serial measurements were performed every hour from a maximum inhalation with the individual in an upright position. The highest value at each measurement was used in the analysis, and each person was his own control. A diurnal variability of < 20% in measured PEF was considered normal.

Test for IgE mediated sensitization

IgE is a type of antibody produced by B-cells in contact with common environmental airborne allergens. The presence of IgE is called allergic sensitization and is a condition that can occur without the individual having symptoms. The term atopy is used when IgE exists in combination with symptoms associated to an allergic disease such as allergic asthma or rhinoconjuctivitis.[155] The most common approach to detect the presence of IgE is by skin prick tests but serum IgE assays are also common.[156] Skin prick tests are safe, cheap and easy to perform, but choosing the allergens to be tested, storing the allergens and interpreting the results can be problematic with this method. Serum specific IgE is a test that is more convenient for the patient, but it is more expensive than the skin prick test.[155] Nowadays, lateral flow tests (LFT) are also available for detection of allergen-specific IgE. They measure antibodies in whole blood, serum or plasma in a shorter time than a skin prick test or a laboratory-based serum IgE assay and the practitioner doesn't need any specific training.[157]

Radioallergosorbent assay (RAST)

There are a number of commercially available antibody-based assays for detecting IgE. The first assay developed for IgE detection was the radioallergosorbent assay (RAST).[158] In recent years, other non-radioactive assay techniques have been developed such as chromogenic enzyme immunoassay (EIA), fluorescence enzyme immunoassay (FEIA) and enzyme-linked immunosorbant assay (ELISA). They are all based on a reaction between the specific IgE in the sample and a bound allergen able to react with a labeled anti-IgE antibody conjugate. Binding of the anti-IgE antibody produces a colour or a light measured with a special photometer and the results are expressed in kilo-units/liter (kU/L) or in specific Ig classes (0–6).[159]

In studies II and III ImmunoCAP Phadiatop (Phadia AB, Uppsala, Sweden) was used as a screening test for IgE-mediated sensitization. A blood sample was taken from each participant during the day of the investigation and the serum was analyzed using an enzyme-linked immunoassay. The serum was tested against a mixture of different common airway allergens such as timothy grass, birch, cats, dogs, and mites. The content of various allergens in the test are updated periodically. The ImmunoCAP Phadiatop is a qualitative test, which means that a positive test result indicates allergic sensitization, but does not provide information regarding which specific allergen the patient reacts to. IgE levels of ≥ 0.35 kU/L were considered to be a positive result.[160]

Lateral flow test (LFT)

The ImmunoCAP Rapid is a LFT that takes 20 minutes to perform. It is a qualitative test designed to measure specific IgE antibodies in the blood. The ImmunoCAP Rapid has one test kit suitable for children and one for adults. Each test kit is able to detect IgE against ten different allergens - airborne allergens in the adult version and a mix of airborne and food allergens in the child test kit.[161] The test kit is disposable and contains developer solution, equipment for capillary blood collection, pipette and an analysis plate. The analysis plate has two wells, one for the blood sample and another for the developer solution. It also contains two test windows with two nitrocellulose membrane strips coated with allergens (5 allergens per window). A control area is also situated in the top of each test window. A positive result is indicated by a pink or red line presented for each allergen in the test windows. A negative result is indicated by absence of colour.[162]

In study IV, ImmunoCAP Rapids adult panel (Phadia AB, Uppsala, Sweden) was used to test for IgE mediated sensitization against ten common European airway allergens: cat, birch, mugwort, timothy, cockroach, dog, olive, wall pellitory, house dust mite, and mould. IgE levels of ≥ 1.0 kU/L were considered to be a positive result.[163] According to the manufacturer, the correspondence between ImmunoCAP Rapid and ImmunoCAP Specific IgE blood tests performed in a lab are as high as 92–98%.[161]

Statistical methods

Data collected in epidemiological studies need to be processed, using descriptive statistics to present them as a clear result, and statistical analyses to generalize them. The variables measured in an epidemiological study can be classified as continuous and categorical, which affects how the results are presented as descriptive statistics, and which methods should be used for their statistical analysis.

In this thesis, the results are estimated using a 95% confidence interval (CI) and a statistical significance level of $p \leq 0.05$. The Statistical Package for Social Sciences (SPSS) for Windows 22.0 was used for all calculations.

Descriptive statistics

The presentation of data comprising continuous variables is usually performed using average values. The most common average values are the arithmetic mean (AM) and the median. The standard deviation (SD) is often used to describe how data are spread around the AM. The standard deviation can be used to calculate the coefficient of variance (CV) to describe the relative dispersion of data around the mean and is often expressed as percentage.

$$CV = 100\frac{\mathrm{SD}}{\mathrm{AM}}$$

Another way to present statistical dispersion is to present the range, the minimum and the maximum value, of a dataset.[164] If the data are log-normally distributed, the geometric mean (GM) and geometric standard deviation (GSD) are more suitable parameters.[165] Categorical variables can be described in percentages in, for example, a frequency table.[164]

Measured personal and stationary air concentrations of trichloramine and THMs (chloroform) were summarized and expressed in AM, SD, GM, GSD and range (min-max). For air concentrations of trichloramine below the LOD, LOD/√2 was used for calculations. For air concentrations of THM below the LOQ, LOQ/√2 was used for calculations.[166] Background variables (demographics) from the spirometry examination (Body Mass Index (BMI)), from the allergy screening and from a questionnaire are summarized for each swimming pool facility. Categorical variables are presented as percentages, and continuous variables as AM, SD, median and range (min-max). Self-reported ocular, nasal and respiratory symptoms are presented individually as percentages. Symptoms reported as having been experienced during the previous week were also categorized as at least one ocular, nasal or respiratory symptom. Measured lung function (FVC and FEV1) and FeNO values 'pre-shift' and in differences between pre-shift and post-shift (abbreviated hereafter to 'change over shift') were expressed in AM, SD and range (min-max).

The daily PEF variability was calculated as the coefficient of variance for each swimming pool worker and presented as a percentage.

Comparison between groups

If the dataset contained different groups, for example an exposed group and a referent group, possible differences between them can be interesting to assess. For continuous data that are normally distributed, an Independent Samples t-test is used, while a non-parametric Mann-Whitney U test is performed for skewed distributions. To compare categorical variables between two groups, a chi-squared test can be appropriate.[164] Differences in measured air concentrations between the different types of facilities were calculated in a Mann-Whitney U test. To assess differences between the groups regarding background variables, a chi-squared test (categorical variables) or a Mann-Whitney U test (continuous variables) was performed.

Correlation and regression analyses

When the individual variables of the dataset have been described, correlation and regression analyzes are often used to see how two or more of them are related. Correlation shows the association between two variables and the strength of the association is represented by the correlation coefficient. Regression is a technique used to predict the value of one variable based on the value of another.[167] Simple linear regression is used in the analysis of continuous variables and is based on one dependent variable (Y) and one independent variable (X). The relationship between the variables is presented as a regression line ($Y = \alpha + \beta X$) where the slope of the line (β) represents the correlation between X and Y. The coefficient of determination (R^2) can also be calculated to determine how much of the variation of Y can be explained by the variation in X. Logistic regression is similar to linear regression except that the dependent variable is dichotomous and the results are expressed as odds ratios (OR). The OR describes the relationship between an exposure and an outcome (disease).[168] Both linear and logistic regression can be multiple, meaning that they include one dependent variable and more than one independent variable (Xs).[167] Linear regression analyses were used to compare personal trichloramine and chloroform concentrations in high-exposure groups with associated stationary measurements in the pool area, predict personal exposure to trichloramine from stationary sampling, and to compare air levels of chloroform and trichloramine.

Differences in self-reported symptoms between the groups and their risk of developing symptoms compared to referents were evaluated via simple logistic regression and multiple logistic regression. A multiple logistic regression was also used to estimate the correlation between reported symptoms and measured personal trichloramine concentrations. In Paper III, the incidence rate ratio (IRR) for developing at least one ocular, nasal or respiratory symptom during the previous week when exposed to a trichloramine concentration above the median in habilitation and rehabilitation swimming pool facilities, were calculated in a Poisson regression with robust error variance. The differences in measured lung function (FVC and FEV1) and FeNO between the groups were calculated via simple and multiple linear regressions. A multiple linear regression also revealed the correlation between lung function and measured personal air concentrations of trichloramine, as well as the correlation between FeNO and personal trichloramine concentrations.

RESULTS

The main findings in Papers I-IV are presented in this section. A more detailed description is presented in each paper.

Demographics

Demographics were collected from all study participants via data from the spirometry examination (BMI) from allergy screening test results, and via a questionnaire. Background variables for 50 referents, 52 conventional swimming pool workers, 23 habilitation and rehabilitation swimming pool workers (one form not completed), and 41 adventure swimming pool workers were registered and are presented for each type of facility in Table 3.

Compared to referents, more of the conventional swimming pool workers reported smoking, self-reported asthma, allergy in childhood, and had a positive allergy screening test. The habilitation and rehabilitation swimming pool workers had a significantly higher average age compared to referents. When adventure swimming pool workers and habilitation and rehabilitation swimming pool workers were compared, they differed significantly according to sex and age.

Table 3. Description of background variables among referents (n = 50), and personnel in: conventional swimming pools (n = 52), habilitation and rehabilitation swimming pools (n = 23), and adventure swimming pools (n = 41).

Background variable		Referents (%)	Conventional workers (%)	Rehabilitation workers (%)	Adventure workers (%)
Sex	Male	15 (30)	16 (31)	3 (13)	17 (41)
	Female	35 (70)	36 (69)	20 (87)	24 (59)
Age (years)	AM ±SD	39.1 ±11.7	36.9 ±12.6	50 ±9	39 ±13
	Median	37.0	33.5	50	38
	min-max	21.0-64.0	21.0-63.0	27-65	19-68
Smoking habits	Smoker	4 (8)	13 (25)	1 (4)	5 (12)
	Ex-smoker	9 (18)	10 (20)	8 (35)	11 (27)
	Never-smoked	37 (74)	28 (55)	14 (61)	25 (61)
BMI (kg/m^2)	AM ±SD	25.0 ±4.2	25.4 ±4.0	25.3 ±4.0	26.0 ±4.2
	Median	24.2	25.3	23.8	25.6
	min-max	18.1-37.7	18.6-39.6	21.0-37.3	19.1-33.5
Time in present job (years)	AM ±SD	7.9 ±7.3	9.1 ±7.7	12.0 ±10.0	10.8 ±10.3
	Median	6.0	7.0	12.5	9.5
	min-max	0.0-31.0	2.0-39.0	1.0-37.0	0.0-35.0
Asthma at any time	Yes	3 (6)	8 (16)	4 (17)	6 (15)
	No	46 (94)	42 (84)	19 (83)	35 (85)
Allergic symptoms during childhood	Yes	16 (36)	24 (50)	4 (18)	18 (46)
	No	28 (64)	24 (50)	18 (82)	21 (54)
Allergy screening[a]	Pos	12 (24)	24 (46)	5 (22)	8 (19)
	Neg	38 (76)	28 (54)	18 (78)	33 (81)

[a]Analyzed with ImmunoCAP Phadiatop among referents, conventional swimming pool workers, and habilitation and rehabilitation swimming pool workers; and with ImmunoCAP Rapid among adventure swimming pool employees.

Abbreviations: n : number of samples; AM : arithmetic mean; SD : standard deviation; BMI : body mass index.

Trichloramine

Trichloramine in air was sampled in all indoor swimming pool facilities included in this thesis: eight conventional facilities (Papers I and II), ten habilitation and rehabilitation facilities (Papers III and IV), and five adventure facilities (Paper IV). AM, SD, GM, GSD and range (min-max) are presented in Table 4 for personal and stationary sampling of trichloramine for each type of facility.

In the conventional facilities, a total of 52 personal and 110 stationary air samples of trichloramine were collected. The mean for personal samples was 71 µg/m^3 and the mean for stationary samples was 170 µg/m^3. The highest personal trichloramine concentration was measured on a swimming schoolteacher (240 µg/m^3). The highest stationary trichloramine concentration in air was measured during a water aerobics session (640 µg/m^3).

In the habilitation and rehabilitation facilities, 24 personal and 35 stationary samples of trichloramine were measured. Due to water contamination and samples destroyed in the analysis, 21 personal and 32 stationary results of trichloramine in air were obtained. Average concentrations of trichloramine were 19 µg/m^3 for personal samples and 23 µg/m^3 for stationary samples. The highest personal trichloramine concentration (76 µg/m^3) was measured on an assistant nurse/rehabilitation assistant and the highest stationary concentration (140 µg/m^3) was registered in the same facility near one of the rehabilitation pools.

In the adventure facilities, 41 personal and 51 stationary samples of trichloramine were measured. Mean air concentrations of trichloramine were 80 µg/m^3 and stationary 183 µg/m^3. The highest personal trichloramine concentration was registered for a lifeguard (410 µg/m^3) and the highest stationary trichloramine concentration was found in the adventure area (850 µg/m^3).

Table 4. Personal and stationary air concentrations of trichloramine in eight conventional, ten habilitation and rehabilitation and five adventure swimming pool facilities.

		Trichloramine ($\mu g/m^3$)					
		n	AM	SD	GM	GSD	min-max
Personal	Conventional	52	71	64	36	4	1-240
	Rehabilitation	21	19	23	8	5	1-76
	Adventure	41	80	82	48	3	1-410
	Total	114	63	67	30	5	1-410
Stationary	Conventional	110	170	130	100	4	1-640
	Rehabilitation	32	23	34	9	4	1-140
	Adventure	51	183	186	106	4	1-850
	Total	193	150	150	70	5	1-850

Abbreviations: *n* : number of samples; AM : arithmetic mean; SD : standard deviation; GM : geometric mean; GSD : geometric standard deviation.

In Paper I, when personal concentrations of trichloramine (Y) measured on the high-exposure group who spent more than 50% of their working day in the conventional swimming pool area were compared with parallel stationary measurements (X) of trichloramine in a linear regression analysis, ($Y= 0.62X - 0.71$) the regression coefficient was significant (95% CI 0.33 – 0.91, $p < 0.001$).

In Paper III, a similar comparison between personal trichloramine exposure (Y) in the high-exposure group in habilitation and rehabilitation swimming pools and stationary sampling (X) was executed. The resulting linear regression analysis ($Y = 0.45X + 3.42$) gave a significant regression coefficient (95% CI 0.31 – 0.59, $p = 0.002$).

Trihalomethanes (THMs)

Trihalomethanes in air were sampled in eight conventional facilities (Paper I), ten habilitation and rehabilitation facilities (Paper III), and in five adventure swimming pool facilities (unpublished). Of all THMs, chloroform was found in the highest concentrations. AM, SD, GM, GSD and range (min-max) are presented in Table 5 for personal and stationary sampling of chloroform for each type of facility.

In the conventional facilities, one personal and one stationary sample were excluded due to water contamination. Mean air concentrations of chloroform were 30 $\mu g/m^3$ for personal sampling ($n = 51$) and 54 $\mu g/m^3$ for stationary sampling ($n = 109$). A swimming teacher / lifeguard were found to have the highest individual concentration of chloroform (170 µg/m^3). The exercise pool in the smallest facility included in the study had the highest stationary chloroform concentration (220 µg/m^3).

In the habilitation and rehabilitation facilities, four personal samples were excluded due to water contamination. Mean air concentrations of chloroform were 15 µg/m^3 for personal sampling (n = 20) and 12 µg/m^3 for stationary sampling (n = 35). The highest personal chloroform concentration (41 µg/m^3) was registered for a rehabilitation assistant. The highest stationary chloroform concentration (45 µg/m^3) was measured near one of the rehabilitation swimming pools.

In the adventure facilities, mean air concentrations of chloroform were 25 µg/m^3 for personal sampling (n = 41) and 35 µg/m^3 for stationary sampling (n = 51). The highest personal chloroform concentration (110 µg/m^3) was measured on a swimming teacher / lifeguard / water aerobics instructor, and the highest stationary concentration (130 µg/m^3) was registered near one of the adventure swimming pools.

Table 5. Personal and stationary air concentrations of chloroform in eight conventional, ten habilitation and rehabilitation, and five adventure swimming pool facilities.

					Chloroform (µg/m^3)		
		n	AM	SD	GM	GSD	min-max
Personal	Conventional	51	30	31	19	3	3-170
	Rehabilitation	20	15	12	9	1	1-41
	Adventure[a]	41	25	24	11	6	1-110
	Total	112	26	27	14	4	1-170
Stationary	Conventional	109	54	49	28	4	1-220
	Rehabilitation	35	12	10	7	1	1-45
	Adventure[a]	51	35	32	14	7	1-130
	Total	195	42	43	19	5	1-220

[a]Chloroform concentrations in air measured in the adventure facilities are not published in any of the papers included in this thesis.

Abbreviations: n : number of samples; AM : arithmetic mean; SD : standard deviation; GM : geometric mean; GSD : geometric standard deviation.

In Paper I, a significant regression coefficient (β 0.56; 95% CI 0.09 – 1.02, p = 0.022) was given when chloroform concentrations in the high-exposure group spending more than 50% of their working day in the conventional swimming pool area were compared to parallel stationary sampling. Chloroform concentrations were found to correlate with trichloramine concentrations in the conventional swimming pool facilities, but not in the habilitation and rehabilitation facilities.

Endotoxins

Endotoxins were measured at 32 different stationary positions in the adventure facilities (Paper IV) with concentrations of < 0.64-25 EU/m^3, where 30 out of 32 samples were below the limit of detection (LOD).

Ocular and respiratory symptoms

Ocular, nasal and respiratory symptoms experienced during the previous week, previous three months and previous year (data not shown) were registered in a questionnaire for referents (n = 50), conventional swimming pool workers (n = 52), habilitation and rehabilitation swimming pool workers (n = 23, one form not completed), and adventure swimming pool workers (n = 41).

In Paper II, the prevalence of self-reported symptoms was compared between conventional swimming pool workers and referents. During the previous week, the exposed group reported a significantly higher prevalence of itchy eyes (36% vs 14%), at least one ocular symptom (62% vs 37%) and blocked nose (55% vs 32%). In a multiple logistic regression (adjusted for smoking, self-reported asthma, allergy in childhood, and IgE mediated sensitization), among the respondents a significantly higher proportion from the exposed group was found to have reported at least one ocular symptom (OR 2.4, 95% CI 1.0-5.7) and at least one nasal symptom (OR 2.3, 95% CI 1.0-5.6). The exposed group were also found to have reported more ocular, nasal, and/or respiratory symptoms during the previous three months (data not shown).

In Paper III, the prevalence of self-reported symptoms was compared between habilitation and rehabilitation swimming pool workers, and referents. A somewhat higher, but not significant, prevalence of symptoms experienced during the previous week and previous three months were recorded for exposed workers compared to referents.

In Paper IV, the prevalence of self-reported symptoms was compared between adventure swimming pool workers, and habilitation and rehabilitation swimming pool workers. A simple logistic regression (unadjusted) revealed a significant difference between the groups regarding self-reported symptoms during the previous week. Compared to the employees in the habilitation and rehabilitation swimming pools, more of the adventure swimming pool workers reported individual symptoms in the form of smarting and itchy eyes, gravel sensation in eyes, dripping and blocked nose, sore throat and dry cough. More of them also reported at least one eye, nose or throat symptom. The significant difference between the groups remained for all symptoms except blocked nose after adjusting for confounding factors (sex and age) in a multiple logistic regression. Prevalence of symptoms during the previous week and comparison between the groups are shown in Table 6. A significantly larger proportion of the adventure swimming pool workers also experienced itchy, burning and irritated eyes during the previous three months, and cough during the previous year compared to habilitation and rehabilitation swimming pool workers (data not shown).

Table 6. Prevalence (%) of ocular, nasal and respiratory symptoms the previous week among workers in habilitation and rehabilitation facilities (n = 23) and adventure facilities (n = 41) and comparison of symptoms between the groups. Bold font indicates statistical significance. The table is adapted from Table 4 in Westerlund et al. 2022.[169]

			Simple logistic regression		Multiple logistic regression[a]	
	Type of facility	Yes (%)	OR	95% CI	OR	95% CI
Any eye symptom	Rehabilitation	9 (39)	1		1	
	Adventure	32 (78)	**5.5**	**1.8-16.9**	**5.6**	**1.5-20.6**
Smarting eyes	Rehabilitation	4 (17)	1		1	
	Adventure	24 (59)	**6.7**	**1.9-23.3**	**6.0**	**1.5-23.5**
Itchy eyes	Rehabilitation	2 (9)	1		1	
	Adventure	17 (42)	**7.4**	**1.5-36.0**	**6.1**	**1.1-33.0**
Gravel sensation in eyes	Rehabilitation	1 (4)	1		1	
	Adventure	15 (37)	**12.7**	**1.6-103.9**	**12.4**	**1.4-111.3**
Any nasal symptom	Rehabilitation	8 (35)	1		1	
	Adventure	27 (66)	**3.6**	**1.2-10.6**	**3.4**	**1.0-11.6**
Dripping nose	Rehabilitation	3 (13)	1		1	
	Adventure	17 (42)	**4.7**	**1.2-18.5**	**4.9**	**1.1-22.1**
Blocked nose	Rehabilitation	6 (21)	1		1	
	Adventure	22 (79)	**3.5**	**1.1-10.6**	3.0	0.9-10.5
Any throat symptom	Rehabilitation	8 (35)	1		1	
	Adventure	27 (66)	**3.6**	**1.2-10.6**	**4.3**	**1.2-15.2**
Sore throat	Rehabilitation	1 (4)	1		1	
	Adventure	15 (37)	**12.7**	**1.6-103.9**	**14.5**	**1.6-132.4**
Dry cough	Rehabilitation	2 (9)	1		1	
	Adventure	14 (34)	**5.4**	**1.1-26.6**	**10.5**	**1.7-64.5**

[a]Adjusted for sex and age.

Abbreviations: n : number of samples; OR : odds ratio; CI : confidence interval.

The prevalence of at least one reported eye, nasal, or throat symptom among referents and personnel working in the different types of swimming pool facilities were compared in a simple logistic regression (unadjusted) and a multiple logistic regression (adjusted for sex and age); results are presented in Table 7. More workers in the conventional and adventure swimming pool facilities reported at least one ('Any') eye symptom compared to referents. In addition, a higher percentage of workers in the adventure pools also reported 'Any throat symptoms' compared to referents.

Table 7. Comparison between referents (*n* = 50) and personnel working in habilitation and rehabilitation facilities (*n* = 23), conventional facilities (*n* = 52), and adventure facilities (*n* = 41) regarding any ocular, nasal and respiratory symptom during the previous week. Bold font indicates statistical significance.

			Simple logistic regression		Multiple logistic regression[a]	
	Type of facility	Yes (%)	OR	95% CI	OR	95% CI
Any eye symptom	Referents	18 (36)	1		1	
	Rehabilitation	9 (39)	1.1	0.4-3.2	1.1	0.4-3.2
	Conventional	32 (62)	**2.8**	**1.3-6.4**	**2.9**	**1.3-6.4**
	Adventure	32 (78)	**6.3**	**2.5-16.2**	**6.4**	**2.5-16.4**
Any nasal symptom	Referents	23 (46)	1		1	
	Rehabilitation	8 (35)	0.6	0.2-1.7	0.6	0.2-1.9
	Conventional	31 (60)	1.7	0.8-3.8	1.7	0.8-3.8
	Adventure	27 (66)	2.3	1.0-5.3	2.2	0.9-5.2
Any throat symptom	Referents	20 (40)	1		1	
	Rehabilitation	8 (35)	0.8	0.3-2.2	0.6	0.2-1.7
	Conventional	26 (50)	1.5	0.7-3.3	1.6	0.7-3.6
	Adventure	27 (66)	**2.9**	**1.2-6.8**	**3.0**	**1.3-7.4**

[a]Adjusted for sex and age.

Abbreviations: *n* : number of samples; OR : odds ratio; CI : confidence interval.

Fraction of exhaled nitric oxide (FeNO)

FeNO concentrations in exhaled air were measured before and after a work shift for referents (*n* = 50), conventional swimming pool workers (*n* = 49, three participants did not complete the examination), habilitation and rehabilitation swimming pool workers (*n* = 23, one participant did not complete the examination), and adventure swimming pool workers (*n* = 41). FeNO values as AM, SD and range (min-max) 'pre-shift' and as 'change over shift' among referents and the three exposed groups of swimming pool workers are presented in Table 8. One referent, three conventional swimming pool workers and two adventure swimming pool workers had FeNO values exceeding the normal range (> 50 ppb).[123]

There were no significant differences in mean concentrations of FeNO in 'pre-shift' or in 'change over shift' between each group of swimming pool workers and referents; but in Paper II, a simple linear regression (unadjusted) revealed a significant difference over shift between the conventional swimming pool workers and referents (+0.7 ppb vs -1.8 ppb; β 2.4 ppb, 95% CI 0.4-4.3: p = 0.013). This significant difference in FeNO over shift was also seen in Paper III in a multiple linear regression (adjusted for asthma and smoking) between habilitation and rehabilitation swimming pool workers and referents (-0.1 ppb vs -1.8 ppb; β 0.27 ppb, 95% CI 0.30-3.67: p = 0.02).

Table 8. Nitric monoxide in exhaled air (FeNO; ppb) 'pre-shift' and 'change over shift' for referents (*n* = 50), conventional swimming pool workers (*n* = 49), habilitation and rehabilitation swimming pool workers (*n* = 23) and adventure swimming pool workers (*n* = 41).

	FeNO (ppb)					
	Pre-shift			Change over shift		
	AM	SD	min-max	AM	SD	min-max
Referents	16.1	10.5	5-67	-1.8	3.7	-16-6
Conventional	18.7	16.5	5-90	+0.7*	5.0	-14-20
Rehabilitation	18.0	6.9	8-41	-0.1*	2.3	-4-4
Adventure	18.7	18.4	2-91	-0.5	7.0	-32-15

Abbreviations: *n* : number of samples; AM : arithmetic mean; SD : standard deviation.

*Significant difference compared to referents (p < 0.05)

Lung function (FVC, FEV1 and PEF)

Spirometry parameters (FVC and FEV1) were measured before and after the work shift for referents (*n* = 50), conventional swimming pool workers (*n* = 52), habilitation and rehabilitation swimming pool workers (*n* = 21, three participants did not complete the examination), and adventure swimming pool workers (*n* = 41). FVC and FEV1 are presented in Table 9 as AM, SD and range (min-max) 'pre-shift' and as 'change over shift' among referents and the three exposed groups of swimming pool workers. Referents and exposed swimming pool workers had lower pre-shift lung function than expected, based on Swedish reference material.[142, 143]

Table 9. Lung function (FVC and FEV1) 'pre-shift' and 'change over shift' for referents (n = 50), conventional swimming pool workers (n = 52), habilitation and rehabilitation swimming pool workers (n = 21) and adventure swimming pool workers (n = 41).

		FVC (%)		FEV1 (%)	
		Pre-shift	Change over shift	Pre-shift	Change over shift
Referents	AM	90.9	0.4	94.0	0.1
	SD	11.8	4.4	12.9	4.2
	min-max	58.0-123.4	-11.2-17.1	61.8-125.6	-12.1-8.9
Conventional	AM	94.3	-0.2	93.8	-0.6
	SD	10.9	3.8	12.4	4.1
	min-max	76.1-117.3	-12.0-16.0	63.7-122.6	-17.7-8.9
Rehabilitation	AM	92.5	0.8	93.7	-0.2
	SD	10.1	3.9	10.8	3.7
	min-max	71.0-108.0	-7.0-13.0	75.0-115.0	-6.0-8.0
Adventure	AM	93.4	-0.7	91.9	-0.4
	SD	12.1	4.0	10.4	4.4
	min-max	63.7-118.0	-16.2-6.2	65.1-113.6	-16.9-9.3

Abbreviations: n : number of samples; AM : arithmetic mean; SD : standard deviation.

The groups were compared in a simple linear regression analysis (unadjusted) according to FVC and FEV1 values 'pre-shift' and in 'change over shift', but no significant difference could be established. In a multiple linear regression analysis (adjusted for IgE mediated sensitization and smoking), the mean 'pre-shift' FEV1/FVC ratios were compared between referents and the different types of swimming pool workers. Compared to referents, the habilitation and rehabilitation swimming pool workers were found to have a lower mean FEV1/FVC ratio (104% vs 101%, $p = 0.27$); the conventional swimming pool workers had a significantly lower ratio (104% vs 100%, $p = 0.04$), as did the adventure swimming pool workers (104% vs 99%, $p = 0.004$).

The habilitation and rehabilitation swimming pool workers (Paper III) performed PEF measurements every hour during their work shift, which resulted in a variance of from 1-5% for each subject.

Trichloramine exposure and adverse health effects

None of the studies included in this thesis were able to, establish an association via regression analysis between measured trichloramine exposure and either self-reported symptoms or measured FeNO and lung function (FVC and FEV1). However, a multiple Poisson regression with robust error variance (adjusted for age and smoking) in Paper III indicated that personal trichloramine exposure above the median (20 $\mu g/m^3$) in habilitation and rehabilitation swimming pools was associated with an increased risk of having developed at least one ocular symptom during the previous week (IRR: 1.2, 95% CI 1.1-1.4). In addition, a Mann-Whitney U test in Paper IV revealed significantly higher median concentrations of trichloramine in the air of adventure swimming pool facilities compared to the habilitation and rehabilitation facilities, for both personal (70 $\mu g/m^3$ vs 8 $\mu g/m^3$, $p < 0.001$) and stationary (150 $\mu g/m^3$ vs 14 $\mu g/m^3$, $p < 0.001$) samples. The adventure swimming pool workers also reported significantly more ocular, nasal and respiratory symptoms.

DISCUSSION

In the studies included in this thesis (Papers I-IV), occupational exposure to trichloramine, THMs and endotoxins in Swedish indoor swimming pool air have been studied and related to adverse health effects. Different types of swimming pool facility offering different kinds of swimming pools and activities have been included.

Trichloramine

The habilitation and rehabilitation swimming pool facilities had the lowest mean concentrations of trichloramine in both personal and stationary samples (19 µg/m^3 and 23 µg/m^3); conventional swimming pools had higher means (71 µg/m^3 and 170 µg/m^3) while the adventure facilities had the highest mean concentrations (80 µg/m^3 and 183 µg/m^3). Median air concentrations of trichloramine found in adventure facilities were also found to be significantly higher than those found in habilitation and rehabilitation facilities, regarding both personal ($p < 0.001$) and stationary ($p < 0.001$) samples. These differences in concentrations between different types of facility are also presented in earlier publications. Studies measuring occupational exposure to trichloramine in conventional swimming pool facilities have found stationary mean concentrations varying between 35-650 µg/m^3 [1, 2, 22, 30, 32, 170-173], while studies in adventure swimming pool facilities measured even higher mean concentrations that varied between 230-1250 µg/m^3.[2, 22, 31, 171] This pattern may be explained by the presence of vigorous water movements and other factors known to effect the trichloramine concentrations in swimming pool air viz. the higher range of water temperatures found in the adventure facilities (conventional facilities: 27-37 °C, habilitation and rehabilitation facilities: 34 °C, and adventure facilities: 27-38 °C).[2, 22] However, despite the fact that the water temperatures in habilitation and rehabilitation swimming pools fall in the higher range, the lowest levels of trichloramine were actually measured in these facilities. Some factors that could explain this might be fewer and smaller pools in these types of facilities, as well as fewer visitors who also engage in calmer activities compared to conventional and adventure pools.

The range of stationary trichloramine concentrations found in the studies included in this thesis (1-850 µg/m^3) is somewhat lower, but still in the same range as earlier published studies measuring occupational trichloramine exposure (20-1920 µg/m^3).[1, 3, 4, 17, 22, 30-32, 170-173] Today's recommended exposure levels for bathers are set as stationary trichloramine levels. The international recommendation is 500 µg/m^3,[5] but individual countries also set their own, such as Sweden (200 µg/m^3),[9] Germany and Switzerland (200 mg/m^3),[6, 7] and France (300 µg/m^3).[8] Because data from personal and stationary measurements differ, the Nordic Expert Group (NEG) have suggested a health based OEL of 100 µg/m^3 for personal measurements and 200 µg/m^3 for stationary measurements.[34] The mean trichloramine concentrations for all personal and stationary measurements found in these studies (63 µg/m^3 and 150 µg/m^3) were below these recommendations. However, 22% of the individual personal trichloramine samples were in the same level or higher than the recommendation from NEG (100 µg/m^3) and 30% of all stationary samples were in the same level or exceeded the recommendations from Sweden, Germany, Switzerland and NEG (200 µg/m^3).

Trihalomethanes (THMs)

The highest concentrations of chloroform, in both personal (170 µg/m^3) and stationary (220 µg/m^3) samples, were found in the conventional swimming pool facilities. Air concentrations of chloroform in indoor swimming pool facilities in earlier studies varied between 8 and 853 µg/m^3.[20, 23, 35, 38, 40, 43, 174-178] Compared to those studies, the measured air concentrations of THMs in the present study (1-220 µg/m^3) were of the same order or lower, and far below the Swedish OEL (10 000 µg/m^3). However, the current OEL may not be suitable for the continuous low-grade exposure occurring in indoor swimming pool facilities. An earlier study associated mean total THM concentrations in air of 205 µg/m^3 and 157 µg/m^3 with an increased cancer risk among elite swimmers and coaches, respectively.[51] However, the Swedish Institute of Environmental medicine has also assessed the risk of cancer associated with swimming in chlorinated swimming pools. They believe that although competitive swimmers experience the highest exposure to chloroform, their cancer risk from it is still negligible.[179]

Air concentrations of chloroform were correlated with air concentrations of trichloramine in the conventional facilities, but not in the habilitation and rehabilitation facilities. The lower air concentrations of both chloroform and trichloramine measured in the habilitation and rehabilitation facilities may be the reason for the lack of correlation.

Notable is the fact that the high humidity in swimming pool facilities has been a problem during analysis as several of the samples could not be analyzed due to water contamination. Although most THM samples could be analyzed, the water affinity of the adsorbent still poses the risk that apparent levels are underestimates.

The relationship between personal and stationary sampling

Earlier studies of occupational exposure in indoor swimming pool air have focused on stationary sampling. The studies included in this thesis are the first that have published data on personal measurements of trichloramine and trihalomethanes. Airborne exposures measured via both personal and stationary sampling do not always give consistent results. Personal measurements generally give higher concentrations than parallel stationary samples, as they are often closer to the source of emission.[180, 181] Personal measurements have usually been found to be 1.2 to 8.5 times higher than samples collected from a stationary position.[182-188] However, the reverse has been found to be the case in the studies included in this thesis concerning the relationship between personal and stationary trichloramine and chloroform concentrations. This might be explained by the fact that the swimming pool employees often have varying work tasks which means that they are not always close to the source of exposure (the swimming pools).

In both conventional swimming pool facilities as well as habilitation and rehabilitation swimming pool facilities, personal trichloramine concentrations from employees spending 50% or more of their working day near the swimming pools (high-exposure group) (Y) were found to be approximately half as high as trichloramine concentrations from parallel stationary sampling (X): (conventional facilities: $Y = 0.62X - 0.71$, $p < 0.001$); habilitation and rehabilitation facilities: $Y = 0.45X + 3.42$, $p = 0.002$). In addition, the linear regression analysis between chloroform concentrations in the high-exposure group in the conventional facilities and parallel stationary sampling gave a significant regression coefficient (β 0.56; 95% CI 0.09 – 1.02, $p = 0.022$).

This relationship between personal and stationary concentrations of trichloramine in swimming pool air has also been noted by the NEG. They have recommended a trichloramine concentration derived from stationary sampling of 200 $\mu g/m^3$, and a health based OEL from personal sampling of 100 $\mu g/m^3$.[34] The recommendation for stationary sampling is based on stationary concentrations of trichloramine associated with self-reported irritation of the eyes and upper respiratory tract among swimming pool workers.[2, 3, 22, 32] Regarding the recommended OEL for personal sampling, the difference between personal and stationary levels presented in this thesis has been taken into account.[189, 190] Based on these findings and present knowledge about occupational exposure and health effects in indoor swimming pool environments, the recommended maximum personal trichloramine concentration from NEG (100 µg/m^3) might be appropriate as a future OEL.

Endotoxins

Endotoxins were measured at 32 different positions in the adventure facilities with concentrations of < 0.64-25 EU/m^3, where 30 out of 32 samples were below the limit of detection (LOD). Measured concentrations were far below the recommended reference value (90 EU/m^3)[69] and lower than the endotoxin levels found in the air of an indoor water park (18-84 µg/m^3).[31] The choice of filters for endotoxin sampling may affect the result as some materials, such as polycarbonate used in this study, are known to absorb endotoxins.[128] An alternative filter material, such as Teflon® or cellulose, may have resulted in higher endotoxin concentrations. However, Dang et al.[31] measured higher endotoxin concentrations in a swimming pool environment even though they used PVC filters, a material also known to absorb endotoxins.

Based on the endotoxin concentrations presented in this thesis, endotoxin exposure in indoor swimming pools does not seem to be an important contributing factor for adverse health effects among swimming pool employees.

Fraction of exhaled nitric oxide (FeNO)

FeNO levels in exhaled air were measured before and after a work shift for swimming pool workers and referents. One referent, three conventional swimming pool workers, and two adventure swimming pool workers had FeNO values higher than the normal range (> 50 ppb),[123] but the mean FeNO levels before a work shift was in the normal range for both swimming pool workers and referents. Mean FeNO levels among swimming pool workers before a shift in the conventional facilities (18.7 ppb), habilitation and rehabilitation facilities (18.0 ppb), and adventure facilities (18.7 ppb) were somewhat, but not significantly, higher compared to referents (16.1 ppb). The mean pre-shift FeNO levels among swimming pool employees were also somewhat higher than mean FeNO levels measured among swimming pool employees in an earlier study (15 ppb)[171] but were more comparable to mean FeNO levels among swimmers in indoor swimming pool facilities (19.7 ppb).[191] It is notable that the conventional and habilitation and rehabilitation swimming pool workers continued to maintain a significantly higher FeNO concentration during the day than did referents. This is an indication of an inflammatory airway response, which was also found in a study of competitive swimmers.[192] Studies examining diurnal variation of FeNO have yielded contradictory results: one found higher FeNO levels in the morning[193] while another study found higher levels in the evening.[194] Thus, it is difficult to comment on how a possible diurnal variation in FeNO affects the results in this thesis.

Lung function (FVC, FEV1 and PEF)

Spirometry was measured (FVC and FEV1) before and after a work shift for swimming pool workers and referents. Both groups had lower lung function than expected compared to the reference material.[142, 143] Nevertheless, lung function was still above the lower limit of normal (LLN), being 80% of the predicted FVC and FEV1. The slightly lower than expected lung function among the participants is considered normal and is unlikely to be linked to exposure in the work environment. No significant differences in FVC or FEV1 between any of the swimming pool groups and referents could be established in either 'pre-shift' samples or in the difference between samples taken before and after a shift ('change over shift'). However, compared to referents (104%) the 'pre-shift' FEV1/FVC ratios were found to be significantly lower among the conventional swimming pool workers (100%, $p = 0.04$), and adventure swimming pool workers (99%, $p = 0.004$), indicating a tendency to airway obstruction in swimming pool employees, especially among those employed in adventure facilities. However, the mean 'pre-shift' FEV1/FVC ratio was above 70% for all of the groups, and cannot therefore be interpreted as airway obstruction.

Among the habilitation and rehabilitation swimming pool workers, variability in PEF during the day was examined. The results (1-5%) did not indicate any abnormal variation that could be associated with any peak exposures.

Trichloramine exposure and adverse health effects

The average trichloramine concentration in stationary samples found among the swimming pool facilities included in this thesis (150 µg/m^3) was lower but still within the same range as those from earlier studies where ocular and upper airway symptoms were reported among swimming pool workers (60-670 µg/m^3).[2, 3, 30-32]

The highest mean trichloramine concentrations were found in the adventure swimming pool facilities (personal: 80 µg/m^3, stationary: 183

μg/m^3) and the second highest mean concentrations were found in the conventional facilities (personal: 71 μg/m^3, stationary: 170 μg/m^3) while the habilitation and rehabilitation swimming pool facilities had the lowest mean concentrations (personal: 19 μg/m^3, stationary: 23 μg/m^3). The same pattern was seen where any (at least one) ocular, nasal or respiratory symptom was reported during the previous week among workers in the different swimming pool facilities compared to referents. The highest increased risk of developing any of these symptoms seems to be associated with workers in adventure swimming pools (Table 7). The lowest FEV1/FVC ratios were also found among adventure swimming pool employees, which might be dependent on the level of trichloramine exposure and a sign of a chronic effect in the airways.

However, linear regression analysis did not show any correlations between measured trichloramine concentrations and self-reported symptoms, lung function, or FeNO. The absence of significant correlations between measured trichloramine concentrations and adverse health effects may be due to different factors. For example, a small sample size can make it hard to detect small differences in symptoms between the groups, and the cross-sectional study design makes it difficult to establish a causal relationship between exposure and health effects. The 'healthy worker' effect may also contribute to a lack of association. The 'healthy worker' effect is based on the fact that those who work are healthier than those who do not work. An explanation for this may be that unhealthy people don't work, or that those experiencing health issues due to work will leave or move to a safer work environment.[195]

In this thesis, adverse health effects have been measured using both subjective and objective methods. Both subjective self-reported health effects and results from the objective lung function examination (FEV1/FVC), and trichloramine exposure have been shown to follow the same pattern, while an association between the objective FeNO measurements and trichloramine exposure is missing. However, the results indicate that there seems to be an increased risk of irritating effects when working in swimming pool facilities, but trichloramine cannot be directly associated with these negative health effects. Thus, it cannot be ruled out that other volatile compounds present in indoor swimming pool air may contribute to, or be the cause of, the negative health effects that have been documented.

CONCLUSIONS

Three different types of swimming pool facilities have been included in this thesis: conventional facilities, habilitation and rehabilitation facilities, and adventure facilities. They represent different types of factors known to affect the air concentrations of trichloramine and trihalomethanes. Chloroform was the dominant THM found in swimming pool air, but the levels were low in comparison with the Swedish OEL. The levels of endotoxins in the air at adventure facilities were also found to be low compared to the reference value. The concentrations of trichloramine found in the air at habilitation and rehabilitation swimming pool facilities were the lowest, while at the adventure facilities they were highest. 22% of personal trichloramine concentrations and 30% of stationary trichloramine concentrations measured at all facilities exceeded recommended OELs presented by NEG (personal: 100 µg/m^3, stationary: 200 µg/m^3).

The adventure facilities had the highest trichloramine levels, as well as the highest prevalence of self-reported ocular and respiratory symptoms, and the lowest FEV1/FVC ratio. The conventional facilities had the second highest trichloramine levels and the second highest prevalence of symptoms as well as the second lowest FEV1/FVC ratio. The habilitation and rehabilitation facilities had the lowest trichloramine exposure; workers in these facilities did not experience as many symptoms as employees in the other types of swimming pools, and they had higher FEV1/FVC ratios. The reference group was considered to be unexposed to trichloramine and thus reported the least ocular and respiratory problems and had the highest FEV1/FVC ratios. In addition, comparisons between conventional swimming pool workers and referents, and between habilitation / rehabilitation swimming pool workers and referents, in the difference in FeNO between samples taken from them before and after a work shift, imply that there may be evidence of a general inflammation of airways among swimming pool workers.

Overall, these findings indicate that there is some kind of irritant or irritants present in indoor swimming pool environments, possibly trichloramine, which causes adverse health effects to the eyes and airways of swimming pool employees. The trichloramine concentrations in personal samples measured among high-exposure workers spending 50% or more of their working day near the swimming pools were also found to be approximately half as high as parallel measurements taken via stationary samples; this is an important indication for a much needed personal-based

Swedish OEL that should be adopted in the future, and which should preferably be in line with the proposal of 100 µg/m^3 made by the NEG.[34]

FUTURE PERSPECTIVES

This thesis provides evidence of the levels of trichloramine and trihalomethanes in the air at different types of Swedish indoor swimming pool facilities to which swimming pool employees are exposed and which may cause adverse health effects. Many studies investigating exposure to DBPs in indoor swimming pool air and adverse health effects among employees and bathers have only collected data at one single point in time. A longitudinal study design, where the exposed swimming pool employees could be followed and traced over a period of time, would be interesting to persue in order to be able to assess the causality between DBP exposure and adverse health effects further.

The levels of chloroform found in swimming pool air and reported in this thesis were considered to be low when compared to the current Swedish OEL. However, the continuous low-level exposure that employees and regular users might experience in swimming pool facilities may require a different type of OEL in order to protect them. The fact that THM uptake occurs via inhalation, through the skin, and by ingestion, needs to be taken into consideration when an individual's total exposure is evaluated. Chloroform in urine and exhaled air can be used as a biomarker to estimate the extent of skin uptake and ingestion. This has been done in some studies, but needs to be evaluated further. The adsorbent used for chloroform sampling in this thesis has an affinity for water, which may have caused problems in the analysis. For further measurements of chloroform in the ambient air of swimming pool facilities, another adsorbent, less sensitive to water should be evaluated.

Another question that still needs to be addressed is: How to control DBP concentrations and prevent them exceeding recommended reference values or OELs. There are many factors known to affect the DBP concentrations in swimming pool air. To prevent the formation of DBPs in swimming pool water, the hygiene of the bathers and type of disinfectant is important. In its new 2021 guidelines on pool baths, the Swedish Public Health Agency has pointed out the importance of the swimming pools providing information about the necessity of bathers being clean and using clean swimwear before swimming in an indoor pool. Different disinfection techniques have not been the focus of this thesis. However, alternative techniques, as well as different complements to chlorination and their effect on the formation and emission of DBPs, are current issues of interest since many new Swedish swimming pool facilities are now being planned and built. Water temperature and water movement (waves, rapids, whirlpools), while affecting the emission of DBPs from water to ambient air, are a key part of the experience the swimming pool facilities want to offer their visitors. To eliminate DBPs that occur in the ambient air of indoor swimming pool facilities, effective ventilation is crucial. Ventilation is another technical aspect, besides disinfection, that is important in the planning of any new indoor swimming pool facility. Evaluating which measures reduce most effectively the levels of DBP exposure in indoor swimming pool facilities is therefore an important topic of immediate interest.

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REFERENCES

1. Jacobs JH, Spaan S, van Rooy GB, et al. Exposure to trichloramine and respiratory symptoms in indoor swimming pool workers. *Eur Respir J*. 2007;29:690-698.
2. Massin N, Bohadana AB, Wild P, Hery M, Toamain JP, Hubert G. Respiratory symptoms and bronchial responsiveness in lifeguards exposed to nitrogen trichloride in indoor swimming pools. *Occup Environ Med*. 1998;55:258-263.
3. Parrat J, Donze G, Iseli C, Perret D, Tomicic C, Schenk O. Assessment of occupational and public exposure to trichloramine in Swiss indoor swimming pools: a proposal for an occupational exposure limit. *Ann Occup Hyg*. 2012;56:264-277.
4. Thickett KM, McCoach JS, Gerber JM, Sadhra S, Burge PS. Occupational asthma caused by chloramines in indoor swimming-pool air. *Eur Respir J*. 2002;19:827-832.
5. WHO. *Guidelines for recreational water environments. Volume 2. Swimming pools and similar environments*. Geneva: World Health Organization; 2006.
6. SIA. *Wasser und Wasseraufbereitungsanlagen in Gemeinschaftsbädern. Anforderungen und ergänzende Bestimmungen für Bau und Betrieb, Schweizer Norm SN 546 385/9*. Zürich, Switzerland: Schweizerischer Ingenieur- und Architektenverein; 2011: 68.
7. Working Group on Indoor Guide Values on the Federal Environment Agency and the States' Health Authorities. *Risk assessment of trichloramine in the air of indoor swimming pools*. Bundesgesundheitsblatt Gesundheitsforschung Gesundheitsschutz; 2011.
8. ANSES. *Évaluation des risques sanitaires liés aux piscines. Partie I : piscines réglementées*. Maisons-Alfort: French Agency for Food, Environmental and Occupational Health & Safety; 2012.
9. Folkhälsomyndigheten (Public Health Agency of Sweden). *Folkhälsomyndighetens allmänna råd om bassängbad HSLF-FS 2021:11*. Stockholm, Sweden: Folkhälsomyndigheten; 2021.
10. Rook JJ. Formation of haloforms during chlorination of natural waters. *Water Treatment and Examination*. 1974;23:234-243.
11. Perrin C, *Nya badhus i många kommuner*, in *Badmästaren*. 2018, Ängelholm: Svenska Badmästareförbundet.
12. Almemark M, Bergström R, Fridén H, et al. *Aktiva badhus*. Stockholm: IVL Svenska Miljöinstitutet; 2015.

13. *Bad och badvatten, riktvärden för bassängvatten,etc. Måttbok måttuppgifter för fritidsanläggningar*. Sveriges Kommuner och Landsting; 2005.
14. Eriksson K, Graff P, Johannesson S, Wastensson G. *Kunskapssammanställning: Kloraminer och exponering i badhus, Rapport 2015:16*. Stockholm: Arbetsmiljöverket; 2015.
15. Gérardina F, Cloteauxa A, Midouxb N. Modeling of variations in nitrogen tri-chloride concentration over time in swimming pool water. *Process Safety and Environmental Protection*. 2015;94:452-462.
16. Cardador MJ, Gallego M. Haloacetic acids in swimming pools: swimmer and worker exposure. *Environ Sci Technol*. 2011;45:5783-90.
17. Weng S, Blatchley ER, 3rd. Disinfection by-product dynamics in a chlorinated, indoor swimming pool under conditions of heavy use: national swimming competition. *Water Res*. 2011;45:5241-8.
18. Zare Afifi M, Blatchley ER, 3rd. Seasonal dynamics of water and air chemistry in an indoor chlorinated swimming pool. *Water Res*. 2015;68:771-83.
19. Erdinger L, Kirsch F, Sonntag HG. Chlorate as an inorganic disinfection by product in swimming pools. *Zentralbl Hyg Umweltmed*. 1999;202:61-75.
20. Aggazzotti G, Fantuzzi G, Righi E, Predieri G. Environmental and biological monitoring of chloroform in indoor swimming pools. *Journal of Chromatography A*. 1995;710:181-190.
21. Aggazzotti G, Fantuzzi G, Tartoni PL, Predieri G. Plasma chloroform concentrations in swimmers using indoor swimming pools. *Archives of Environmental Health*. 1990;45:175-179.
22. Hery M, Hecht G, Gerber JM, Gendre JC, Hubert G. Exposure to chloramines in the atmosphere of indoor swimming pools. *Ann Occup Hyg*. 1995;39:427-439.
23. Nitter TB, Svendsen KVH. Determinants of Exposure and Variability of Trihalomethanes in the Air of Three Indoor Swimming Pools. *Ann Work Expo Health*. 2019;63:560-567.
24. Hailin G, Wallace GG, O'Halloran RAJ. Determination of trace amounts of chloramines by liquid chromatographic separation and amperometric detection. *Analyt Chim Acta*. 1990;237:149-153.
25. Holzwarth G, Balmer RG, Soni L. The fate of chlorine and chloramines in cooling towers. *Water Res*. 1984;18:1421-1427.
26. Bonvallot N, Glorennec P, Zmirou D. Derivation of a toxicity reference value for nitrogen trichloride as a disinfection by-product. *Regul Toxicol Pharmacol*. 2010;56:357-64.

27. Barbee SJ, Thackara JW, Rinehart WE. Acute inhalation toxicology of nitrogen trichloride. *American Industrial Hygiene Association Journal*. 1983;44:145-6.
28. Gagnaire F, Azim S, Bonnet P, Hecht G, Hery M. Comparison of the sensory irritation response in mice to chlorine and nitrogen trichloride. *Journal of Applied Toxicology*. 1994;14:405-409.
29. Carbonnelle S, Francaux M, Doyle I, et al. Changes in serum pneumoproteins caused by short-term exposures to nitrogen trichloride in indoor chlorinated swimming pools. *Biomarkers*. 2002;7:464-78.
30. Chu TS, Cheng SF, Wang GS, Tsai SW. Occupational exposures of airborne trichloramine at indoor swimming pools in Taipei. *Sci Total Environ*. 2013;461-462:317-322.
31. Dang B, Chen L, Mueller C, et al. Ocular and respiratory symptoms among lifeguards at a hotel indoor waterpark resort. *J Occup Environ Med*. 2010;52:207-213.
32. Fantuzzi G, Righi E, Predieri G, Giacobazzi P, Petra B, Aggazzotti G. Airborne trichloramine (NCl(3)) levels and self-reported health symptoms in indoor swimming pool workers: dose-response relationships. *J Expo Sci Environ Epidemiol*. 2013;23:88-93.
33. Nordberg GF, Lundstrom NG, Forsberg B, et al. Lung function in volunteers before and after exposure to trichloramine in indoor pool environments and asthma in a cohort of pool workers. *BMJ Open*. 2012;2.
34. Wastensson G, Eriksson K. 152. Inorganic chloramines *Work and Health*. 2019;53(2).
35. Lahl U, Bätjer K, Düszeln Jv, Gabel B, Stachel B, Thiemann W. Distribution and balance of vaolatile halogenated hydrocarbons in the water and air of covered swimming pools using chlorine for water disinfiction. *Water Res*. 1981;15:803-814.
36. Manasfi T, Temime-Roussel B, Coulomb B, Vassalo L, Boudenne JL. Occurrence of brominated disinfection byproducts in the air and water of chlorinated seawater swimming pools. *Int J Hyg Environ Health*. 2017;220:583-590.
37. Parinet J, Tabaries S, Coulomb B, Vassalo L, Boudenne JL. Exposure levels to brominated compounds in seawater swimming pools treated with chlorine. *Water Res*. 2012;46:828-36.
38. Aggazzotti G, Fantuzzi G, Righi E, Predieri G. Blood and breath analyses as biological indicators of exposure to trihalomethanes in indoor swimming pools. *Science of the Total Environment*. 1998;217:155-63.
39. Aggazzotti G, Fantuzzi G, Righi E, Tartoni P, Cassinadri T, Predieri G. Chloroform in alveolar air of individuals attending

indoor swimming pools. *Archives of Environmental Health*. 1993;48:250-4.

40. Fantuzzi G, Righi E, Predieri G, Ceppelli G, Gobba F, Aggazzotti G. Occupational exposure to trihalomethanes in indoor swimming pools. *Sci Total Environ*. 2001;264:257-65.
41. Marco E, Lourencetti C, Grimalt JO, et al. Influence of physical activity in the intake of trihalomethanes in indoor swimming pools. *Environ Res*. 2015;140:292-9.
42. Aprea MC, Banchi B, Liana Lunghini, Pagliantini M, Peruzzi A, Sciarra G. Disinfection of swimming pools with chlorine and derivatives: formation of organochlorinated and organobrominated compounds and exposure of pool personnel and swimmers. *Natural Science*. 2010;2:68-78.
43. Erdinger L, Kuhn KP, Kirsch F, et al. Pathways of trihalomethane uptake in swimming pools. *Int J Hyg Environ Health*. 2004;207:571-575.
44. Levesque B, Ayotte P, LeBlanc A, et al. Evaluation of dermal and respiratory chloroform exposure in humans. *Environmental Health Perspectives*. 1994;102:1082-7.
45. Lee J, Ha KT, Zoh KD. Characteristics of trihalomethane (THM) production and associated health risk assessment in swimming pool waters treated with different disinfection methods. *Sci Total Environ*. 2009;407:1990-7.
46. Chen MJ, Lin CH, Duh JM, Chou WS, Hsu HT. Development of a multi-pathway probabilistic health risk assessment model for swimmers exposed to chloroform in indoor swimming pools. *J Hazard Mater*. 2011;185:1037-44.
47. Florentin A, Hautemanière A, Hartemann P. Health effects of disinfection by-products in chlorinated swimming pools. *Int J Hyg Environ Health*. 2011;214:461-9.
48. Hang C, Zhang B, Gong T, Xian Q. Occurrence and health risk assessment of halogenated disinfection byproducts in indoor swimming pool water. *Sci Total Environ*. 2016;543:425-431.
49. Panyakapo M, Soontornchai S, Paopuree P. Cancer risk assessment from exposure to trihalomethanes in tap water and swimming pool water. *J Environ Sci (China)*. 2008;20:372-8.
50. Villanueva CM, Cantor KP, Grimalt JO, et al. Bladder cancer and exposure to water disinfection by-products through ingestion, bathing, showering, and swimming in pools. *American Journal of Epidemiology*. 2007;165:148-56.
51. Gouveia P, Felgueiras F, Mourão Z, Fernandes EO, Moreira A, Gabriel MF. Predicting health risk from exposure to trihalomethanes in an Olympic-size indoor swimming pool among

elite swimmers and coaches. *J Toxicol Environ Health A*. 2019;82:577-590.
52. SWEA. *Hygieniska gränsvärden AFS 2018:1*. Stockholm: Swedish Work Environment Authority; 2018.
53. *Endotoxins Health-based recommended occupational exposure limit*. The Hague: Dutch expert Committee on Occupational Safety a Committee of the Health Council of the Netherlands in cooperation with the Nordic Expert Group for Criteria Documentation of Health Risks from Chemicals; 2010.
54. Hasday JD, Bascom R, Costa JJ, Fitzgerald T, Dubin W. Bacterial endotoxin is an active component of cigarette smoke. *Chest*. 1999;115:829-35.
55. Kullman GJ, Thorne PS, Waldron PF, et al. Organic dust exposures from work in dairy barns. *Am Ind Hyg Assoc J*. 1998;59:403-413.
56. Laitinen S, Kangas J, Husman K, Susitaival P. Evaluation of exposure to airborne bacterial endotoxins and peptidoglycans in selected work environments. *Ann Agric Environ Med*. 2001;8:213-219.
57. Simpson JC, Niven RM, Pickering CA, Oldham LA, Fletcher AM, Francis HC. Comparative personal exposures to organic dusts and endotoxin. *Ann Occup Hyg*. 1999;43:107-115.
58. Keman S, Jetten M, Douwes J, Borm PJ. Longitudinal changes in inflammatory markers in nasal lavage of cotton workers. Relation to endotoxin exposure and lung function changes. *Int Arch Occup Environ Health*. 1998;71:131-137.
59. Buchan RM, Rijal P, Sandfort D, Keefe T. Evaluation of airborne dust and endotoxin in corn storage and processing facilities in Colorado. *Int J Occup Med Environ Health*. 2002;15:57-64.
60. Alwis KU, Mandryk J, Hocking AD. Exposure to biohazards in wood dust: bacteria, fungi, endotoxins, and (1-->3)-beta-D-glucans. *Appl Occup Environ Hyg*. 1999;14:598-608.
61. Dutkiewicz J, Krysińska-Traczyk E, Prazmo Z, Skoŕska C, Sitkowska J. Exposure to airborne microorganisms in Polish sawmills. *Ann Agric Environ Med*. 2001;8:71-80.
62. Mandryk J, Alwis KU, Hocking AD. Effects of personal exposures on pulmonary function and work-related symptoms among sawmill workers. *Ann Occup Hyg*. 2000;44:281-289.
63. Prazmo Z, Dutkiewicz J, Skórska C, Sitkowska J, Cholewa G. Exposure to airborne Gram-negative bacteria, dust and endotoxin in paper factories. *Ann Agric Environ Med*. 2003;10:93-100.

64. Rose CS, Martyny JW, Newman LS, et al. "Lifeguard lung": endemic granulomatous pneumonitis in an indoor swimming pool. *Am J Public Health*. 1998;88:1795-800.
65. Heldal KK, Madsø L, Eduard W. Airway inflammation among compost workers exposed to actinomycetes spores. *Ann Agric Environ Med*. 2015;22:253-8.
66. Schlünssen V, Madsen AM, Skov S, Sigsgaard T. Does the use of biofuels affect respiratory health among male Danish energy plant workers? *Occup Environ Med*. 2011;68:467-73.
67. Michel O, Dentener M, Corazza F, Buurman W, Rylander R. Healthy subjects express differences in clinical responses to inhaled lipopolysaccharide that are related with inflammation and with atopy. *J Allergy Clin Immunol*. 2001;107:797-804.
68. Sandström T, Bjermer L, Rylander R. Lipopolysaccharide (LPS) inhalation in healthy subjects increases neutrophils, lymphocytes and fibronectin levels in bronchoalveolar lavage fluid. *Eur Respir J*. 1992;5:992-6.
69. The Nordic Expert Group for Criteria Documentation of Health Risks from Chemicals and the Dutch Expert Committee on Occupational Safety. *144. Endotoxins*. Gothenburg, Sweden: The Nordic Expert Group for Criteria Documentation of Health Risks from Chemicals; 2011.
70. Castellan RM, Olenchock SA, Kinsley KB, Hankinson JL. Inhaled endotoxin and decreased spirometric values. An exposure-response relation for cotton dust. *N Engl J Med*. 1987;317:605-610.
71. Post W, Heederik D, Houba R. Decline in lung function related to exposure and selection processes among workers in the grain processing and animal feed industry. *Occup Environ Med*. 1998;55:349-55.
72. Smid T, Heederik D, Houba R, Quanjer PH. Dust- and endotoxin-related acute lung function changes and work-related symptoms in workers in the animal feed industry. *Am J Ind Med*. 1994;25:877-88.
73. Farokhi A, Heederik D, Smit LAM. Respiratory health effects of exposure to low levels of airborne endotoxin - a systematic review. *Environ Health*. 2018;17:14.
74. International Occupational Hygiene Association. *Occupational Hygiene - What is Occupational Hygiene? [Internet]*. 2022; Available from: https://www.ioha.net/about/occupational-hygiene/.
75. Rappaport SM, Kupper LL. *Quantitative Exposure Assessment*. El Cerrito, California, USA: Stephen Rappaport; 2008.
76. Mulhausen JR, Damiano J. *A Strategy for Assessing and Managing Occupational Exposures* 2ed., Fairfax: American Industrial

Hygiene Association Exposure Assessment Strategies Committee; 1998.
77. *SS-EN 689:2018+AC:2019 Workplace exposure - Measurement of exposure by inhalation to chemical agents - Strategy for testing compliance with occupational exposure limit values*. Stockholm: Swedish Standards Institution; 2019.
78. Arbetsmedicinska institutet. *Rekommenderade hygieniska gränsvärden för luftföroreningar på arbetsplatsen, AI-rapport nr 13.*, Stockholm: Arbetsmedicinska institutet; 1969.
79. Schenk L. *Hygieniska gränsvärden – till vilken nytta?* , Stockholm: US-AB Universitetsservice AB; 2013.
80. Johansson G, Schenk L. *Kunskapsöversikt: Reach och hygieniska gränsvärden, Rapport 2010:5*. Stockholm: Arbetsmiljöverket; 2010.
81. Edling C, Nordberg G, Albin M, Nordberg M. Arbets- och miljömedicin - begrepp och trender. In. Edling C, et al., Editors *Arbets- och miljömedicin - en lärobok om hälsa och miljö*. Lund: Studentlitteratur; 2010.
82. Brüning T, Bartsch R, Bolt HM, et al. Sensory irritation as a basis for setting occupational exposure limits. *Arch Toxicol*. 2014;88:1855-79.
83. Rolando M, Zierhut M. The ocular surface and tear film and their dysfunction in dry eye disease. *Surv Ophthalmol*. 2001;45 Suppl 2:S203-10.
84. Wolkoff P, Skov P, Franck C, Petersen LN. Eye irritation and environmental factors in the office environment--hypotheses, causes and a physiological model. *Scand J Work Environ Health*. 2003;29:411-30.
85. Doty RL, Cometto-Muñiz JE, Jalowayski AA, Dalton P, Kendal-Reed M, Hodgson M. Assessment of upper respiratory tract and ocular irritative effects of volatile chemicals in humans. *Crit Rev Toxicol*. 2004;34:85-142.
86. Arts JH, Rennen MA, de Heer C. Inhaled formaldehyde: evaluation of sensory irritation in relation to carcinogenicity. *Regul Toxicol Pharmacol*. 2006;44:144-60.
87. Lang I, Bruckner T, Triebig G. Formaldehyde and chemosensory irritation in humans: a controlled human exposure study. *Regul Toxicol Pharmacol*. 2008;50:23-36.
88. Kjaergaar SK, Hodgson M. The assessment of irritation using clinical methods and questionnaires. *Aihaj*. 2001;62:711-6.
89. Koeppen BM, Stanton BA. Introduction to the respiratory system. In: *Berne and Levy Physiology*. 2018.

90. Moscato G, Vandenplas O, Gerth Van Wijk R, et al. Occupational rhinitis. *Allergy*. 2008;63:969-80.
91. Beard S. Rhinitis. *Prim Care*. 2014;41:33-46.
92. Bjermer L, Sandström T. Diagnostik och monitorering av astma In: *Lungmedicin.* Sandström T, Eklund A; 2015.
93. Global burden of 369 diseases and injuries in 204 countries and territories, 1990-2019: a systematic analysis for the Global Burden of Disease Study 2019. *Lancet*. 2020;396:1204-1222.
94. Balmes J, Becklake M, Blanc P, et al. American Thoracic Society Statement: Occupational contribution to the burden of airway disease. *Am J Respir Crit Care Med*. 2003;167:787-97.
95. Cakir E, Ersu R, Uyan ZS, et al. The prevalence and risk factors of asthma and allergic diseases among working adolescents. *Asian Pac J Allergy Immunol*. 2010;28:122-9.
96. Espuga M, Muñoz X, Plana E, et al. Prevalence of possible occupational asthma in hairdressers working in hair salons for women. *Int Arch Allergy Immunol*. 2011;155:379-88.
97. Singh T, Bello B, Jeebhay MF. Risk factors associated with asthma phenotypes in dental healthcare workers. *Am J Ind Med*. 2013;56:90-9.
98. Vandenplas O. Occupational asthma: etiologies and risk factors. *Allergy Asthma Immunol Res*. 2011;3:157-67.
99. Quirce S, Campo P, Domínguez-Ortega J, et al. New developments in work-related asthma. *Expert Rev Clin Immunol*. 2017;13:271-281.
100. Breton CV, Zhang Z, Hunt PR, Pechter E, Davis L. Characteristics of work related asthma: results from a population based survey. *Occup Environ Med*. 2006;63:411-5.
101. Henneberger PK, Derk SJ, Sama SR, et al. The frequency of workplace exacerbation among health maintenance organisation members with asthma. *Occup Environ Med*. 2006;63:551-7.
102. Brant A. Baker's asthma. *Curr Opin Allergy Clin Immunol*. 2007;7:152-5.
103. Mazurek JM, Weissman DN. Occupational Respiratory Allergic Diseases in Healthcare Workers. *Curr Allergy Asthma Rep*. 2016;16:77.
104. Cormier M, Lemière C. Occupational asthma. *Int J Tuberc Lung Dis*. 2020;24:8-21.
105. Friedman-Jimenez G, Harrison D, Luo H. Occupational asthma and work-exacerbated asthma. *Semin Respir Crit Care Med*. 2015;36:388-407.
106. Vandenplas O, Suojalehto H, Cullinan P. Diagnosing occupational asthma. *Clin Exp Allergy*. 2017;47:6-18.

107. Vandenplas O, Godet J, Hurdubaea L, et al. Are high- and low-molecular-weight sensitizing agents associated with different clinical phenotypes of occupational asthma? *Allergy*. 2019;74:261-272.
108. Merget R, Caspari C, Dierkes-Globisch A, et al. Effectiveness of a medical surveillance program for the prevention of occupational asthma caused by platinum salts: a nested case-control study. *J Allergy Clin Immunol*. 2001;107:707-12.
109. Merget R, Sander I, van Kampen V, et al. Occupational immediate-type asthma and rhinitis due to rhodium salts. *Am J Ind Med*. 2010;53:42-6.
110. Wittczak T, Dudek W, Walusiak-Skorupa J, et al. Metal-induced asthma and chest X-ray changes in welders. *Int J Occup Med Environ Health*. 2012;25:242-50.
111. Blomqvist A, Düzakin-Nystedt M, Ohlson CG, et al. Airways symptoms, immunological response and exposure in powder painting. *Int Arch Occup Environ Health*. 2005;78:123-31.
112. Nilsson R, Nordlinder R, Wass U, Meding B, Belin L. Asthma, rhinitis, and dermatitis in workers exposed to reactive dyes. *Br J Ind Med*. 1993;50:65-70.
113. Moscato G, Pala G, Perfetti L, Frascaroli M, Pignatti P. Clinical and inflammatory features of occupational asthma caused by persulphate salts in comparison with asthma associated with occupational rhinitis. *Allergy*. 2010;65:784-90.
114. Pala G, Pignatti P, Perfetti L, et al. Occupational rhinitis and asthma due to cabreuva wood dust. *Ann Allergy Asthma Immunol*. 2010;104:268-9.
115. Budnik LT, Preisser AM, Permentier H, Baur X. Is specific IgE antibody analysis feasible for the diagnosis of methylenediphenyl diisocyanate-induced occupational asthma? *Int Arch Occup Environ Health*. 2013;86:417-30.
116. Merget R, Kulzer R, Dierkes-Globisch A, et al. Exposure-effect relationship of platinum salt allergy in a catalyst production plant: conclusions from a 5-year prospective cohort study. *J Allergy Clin Immunol*. 2000;105:364-70.
117. Zeiss CR, Patterson R, Pruzansky JJ, Miller MM, Rosenberg M, Levitz D. Trimellitic anhydride-induced airway syndromes: clinical and immunologic studies. *J Allergy Clin Immunol*. 1977;60:96-103.
118. Castano R, Thériault G. Defining and classifying occupational rhinitis. *J Laryngol Otol*. 2006;120:812-7.
119. Storaas T, Steinsvåg SK, Florvaag E, Irgens A, Aasen TB. Occupational rhinitis: diagnostic criteria, relation to lower airway

symptoms and IgE sensitization in bakery workers. *Acta Otolaryngol*. 2005;125:1211-7.

120. Rothman KJ. *Epidemiology - An Introduction*. New York: Oxford University Press, Inc.; 2012.
121. Ahlbom A, Norell S. *Grunderna i Epidemiologi*. 2 ed., Lund: Studentlitteratur; 1987.
122. World Medical Association Declaration of Helsinki: ethical principles for medical research involving human subjects. *Jama*. 2013;310:2191-4.
123. Dweik RA, Boggs PB, Erzurum SC, et al. An official ATS clinical practice guideline: interpretation of exhaled nitric oxide levels (FENO) for clinical applications. *Am J Respir Crit Care Med*. 2011;184:602-615.
124. Michalski R. Application of ion chromatography in clinical studies and pharmaceutical industry. *Mini Rev Med Chem*. 2014;14:862-72.
125. Harris DC. *Quantitative Chemical Analysis*. 6 ed., New York: W. H. Freeman and Company; 2003.
126. US Environmental Protection Agency. *Method TO-17: Determination of volatile organic compounds in ambient air using active sampling onto sorbent tubes*. 2nd ed., Cincinnati: U.S. EPA; 1999.
127. SUPELCO och Perkin Elmer™. *Users guide for prepacked sample tubes*.
128. *SS-EN 14031:2021 Workplace exposure - Quantitative measurement of airborne endotoxins*. Stockholm: Swedish Standards Institution; 2021.
129. Duquenne P, Marchand G, Duchaine C. Measurement of endotoxins in bioaerosols at workplace: a critical review of literature and a standardization issue. *Ann Occup Hyg*. 2013;57:137-72.
130. Story DA, Tait AR. Survey Research. *Anesthesiology*. 2019;130:192-202.
131. Nieuwenhuijsen MJ. Design of exposure questionnaires for epidemiological studies. *Occup Environ Med*. 2005;62:272-80, 212-4.
132. Infante-Rivard C, Jacques L. Empirical study of parental recall bias. *Am J Epidemiol*. 2000;152:480-6.
133. Nielsen J, Welinder H, Jonsson B, Axmon A, Rylander L, Skerfving S. Exposure to hexahydrophthalic and methylhexahydrophthalic anhydrides--dose-response for sensitization and airway effects. *Scand J Work Environ Health*. 2001;27:327-334.

134. Reijula K, Sundman-Digert C. Assessment of indoor air problems at work with a questionnaire. *Occup Environ Med*. 2004;61:33-8.
135. Wålinder R, Norbäck D, Wieslander G, Smedje G, Erwall C, Venge P. Nasal patency and biomarkers in nasal lavage--the significance of air exchange rate and type of ventilation in schools. *Int Arch Occup Environ Health*. 1998;71:479-86.
136. Nathan C, Xie QW. Nitric oxide synthases: roles, tolls, and controls. *Cell*. 1994;78:915-8.
137. Dweik RA, Comhair SA, Gaston B, et al. NO chemical events in the human airway during the immediate and late antigen-induced asthmatic response. *Proc Natl Acad Sci U S A*. 2001;98:2622-7.
138. Olin AC. Induced sputum, exhaled nitric oxide, and particles in exhaled air in assessing airways inflammation in occupational exposures. *Clin Chest Med*. 2012;33:771-82.
139. Miller MR, Hankinson J, Brusasco V, et al. Standardisation of spirometry. *Eur Respir J*. 2005;26:319-338.
140. Graham BL, Steenbruggen I, Miller MR, et al. Standardization of Spirometry 2019 Update. An Official American Thoracic Society and European Respiratory Society Technical Statement. *Am J Respir Crit Care Med*. 2019;200:e70-e88.
141. Quanjer PH, Tammeling GJ, Cotes JE, Pedersen OF, Peslin R, Yernault JC. Lung volumes and forced ventilatory flows. Report Working Party Standardization of Lung Function Tests, European Community for Steel and Coal. Official Statement of the European Respiratory Society. *Eur Respir J Suppl*. 1993;16:5-40.
142. Hedenström H, Malmberg P, Agarwal K. Reference values for lung function tests in females. Regression equations with smoking variables. *Bull Eur Physiopathol Respir*. 1985;21:551-557.
143. Hedenström H, Malmberg P, Fridriksson HV. Reference values for lung function tests in men: regression equations with smoking variables. *Upsala J Med Sci*. 1986;91:299-310.
144. Quanjer PH, Stanojevic S, Cole TJ, et al. Multi-ethnic reference values for spirometry for the 3-95-yr age range: the global lung function 2012 equations. *Eur Respir J*. 2012;40:1324-43.
145. Miller MR, Quanjer PH, Swanney MP, Ruppel G, Enright PL. Interpreting lung function data using 80% predicted and fixed thresholds misclassifies more than 20% of patients. *Chest*. 2011;139:52-9.
146. Pellegrino R, Viegi G, Brusasco V, et al. Interpretative strategies for lung function tests. *Eur Respir J*. 2005;26:948-68.
147. Leroyer C, Perfetti L, Trudeau C, L'Archevěque J, Chan-Yeung M, Malo JL. Comparison of serial monitoring of peak expiratory flow

and FEV1 in the diagnosis of occupational asthma. *Am J Respir Crit Care Med*. 1998;158:827-32.
148. Nicholson PJ, Cullinan P, Taylor AJ, Burge PS, Boyle C. Evidence based guidelines for the prevention, identification, and management of occupational asthma. *Occup Environ Med*. 2005;62:290-9.
149. Casale R, Pasqualetti P. Cosinor analysis of circadian peak expiratory flow variability in normal subjects, passive smokers, heavy smokers, patients with chronic obstructive pulmonary disease and patients with interstitial lung disease. *Respiration*. 1997;64:251-6.
150. Goyal M, Goel A, Kumar P, et al. Circadian rhythm of peak expiratory flow rate in healthy north Indian men. *Indian J Physiol Pharmacol*. 2008;52:64-8.
151. Goel A, Goyal M, Singh R, Verma N, Tiwari S. Diurnal Variation in Peak Expiratory Flow and Forced Expiratory Volume. *J Clin Diagn Res*. 2015;9:Cc05-7.
152. Quackenboss JJ, Lebowitz MD, Krzyzanowski M. The normal range of diurnal changes in peak expiratory flow rates. Relationship to symptoms and respiratory disease. *Am Rev Respir Dis*. 1991;143:323-30.
153. Hayati F, Maghsoodloo S, DeVivo MJ, Thomas RE, Lemiere C. Quality control chart method for analyzing PEF variability in occupational asthma. *Am J Ind Med*. 2008;51:223-8.
154. Burge CB, Moore VC, Pantin CF, Robertson AS, Burge PS. Diagnosis of occupational asthma from time point differences in serial PEF measurements. *Thorax*. 2009;64:1032-6.
155. Ansotegui IJ, Melioli G, Canonica GW, et al. IgE allergy diagnostics and other relevant tests in allergy, a World Allergy Organization position paper. *World Allergy Organ J*. 2020;13:100080.
156. Johansson SG, Hourihane JO, Bousquet J, et al. A revised nomenclature for allergy. An EAACI position statement from the EAACI nomenclature task force. *Allergy*. 2001;56:813-24.
157. Chapman MD. Lateral flow tests for allergy diagnosis: point-of-care or point of contention? *Int Arch Allergy Immunol*. 2010;152:301-2.
158. Wide L, Bennich H, Johansson SG. Diagnosis of allergy by an in-vitro test for allergen antibodies. *Lancet*. 1967;2:1105-7.
159. Wachholz PA, Dearman RJ, Kimber I. Detection of Allergen-Specific IgE Antibody Responses. *J Immunotoxicol*. 2005;1:189-99.

160. *ImmunoCAP Phadiatop Bruksanvisning 52-5205-SE/15*. Uppsala: Phadia AB; 2013.
161. *ImmunoCAP Rapid Broschyr A5*. Thermo Fischer Scientific; 2016.
162. Diaz-Vazquez C, Torregrosa-Bertet MJ, Carvajal-Urueña I, et al. Accuracy of ImmunoCAP Rapid in the diagnosis of allergic sensitization in children between 1 and 14 years with recurrent wheezing: the IReNE study. *Pediatr Allergy Immunol*. 2009;20:601-9.
163. Johansson A-S. *Performance ImmunoCAP Rapid*. Thermo Fisher; 2020.
164. Ejlertsson G. *Statistik för hälsovetenskaperna*. Lund: Studentlitteratur; 2003.
165. Seixas NS, Robins TG, Moulton LH. The use of geometric and arithmetic mean exposures in occupational epidemiology. *Am J Ind Med*. 1988;14:465-77.
166. Hornung RW, Reed LD. Estimation of avarage concentration in the presence of nondetectable values. *Appl Occup Environ Hyg*. 1990;5:46-51.
167. Campbell M, Machin D, Walters S. *Medical statistics: a textbook for the health sciences*. Chichester: Wiley; 2007.
168. Bring J, Taube A, Wikman P. *Introduktion till medicinsk statistik*. Lund: Studentlitteratur; 2015.
169. Westerlund J, Bryngelsson IL, Fornander L, Löfstedt H, Graff P. Occupational Exposure to Trichloramine and Endotoxins: Adverse Health Effects Among Personnel in Adventure and Rehabilitation Swimming Pool Facilities. *J Occup Environ Med*. 2022.
170. Lévesque B, Vézina L, Gauvin D, Leroux P. Investigation of Air Quality Problems in an Indoor Swimming Pool: A Case Study. *Ann Occup Hyg*. 2015;59:1085-9.
171. Fornander L, Ghafouri B, Lindahl M, Graff P. Airway irritation among indoor swimming pool personnel: trichloramine exposure, exhaled NO and protein profiling of nasal lavage fluids. *Int Arch Occup Environ Health*. 2013;86:571-580.
172. Catto C, Sabrina S, Ginette CT, Manuel R, Robert T. Occurrence and spatial and temporal variations of disinfection by-products in the water and air of two indoor swimming pools. *Int J Environ Res Public Health*. 2012;9:2562-86.
173. Bessonneau V, Derbez M, Clement M, Thomas O. Determinants of chlorination by-products in indoor swimming pools. *Int J Hyg Environ Health*. 2011;215:76-85.
174. Cammann K, Hubner K. Trihalomethane concentrations in swimmers' and bath attendants' blood and urine after swimming

or working in indoor swimming pools. *Archives of Environmental Health*. 1995;50:61-5.

175. Matthiessen A, Jentsch F. Trihalomethanes in air of indoor swimming pools and uptake by swimmers. In. G Raw, Aizlewood C, and Warren P, Editors *Indoor Air 99: proceedings of the 8th International Conference on Indoor Air Quality and Climate 1999*. Edinburgh; 1999:448-453.
176. Tardif R, Catto C, Haddad S, Simard S, Rodriguez M. Assessment of air and water contamination by disinfection by-products at 41 indoor swimming pools. *Environ Res*. 2016;148:411-420.
177. Caro J, Gallego M. Alveolar air and urine analyses as biomarkers of exposure to trihalomethanes in an indoor swimming pool. *Environ Sci Technol*. 2008;42:5002-7.
178. Lévesque B, Ayotte P, Tardif R, et al. Evaluation of the health risk associated with exposure to chloroform in indoor swimming pools. *J Toxicol Environ Health A*. 2000;61:225-43.
179. Stamyr K, Johanson G, *Hälsoriskbedömning av trihalometaner i bassängbad*. 2006, Karolinska Institutet - Institutet för miljömedicin: Stockholm.
180. Leung PL, Harrison RM. Evaluation of personal exposure to monoaromatic hydrocarbons. *Occup Environ Med*. 1998;55:249-57.
181. Lange JH, Kuhn BD, Thomulka KW. A study of area and personal airborn asbestos samples during abatement in a crawl space. *Indoor and Built Environment*. 2000;9:192-200.
182. Botham RA, Hughson G, Parker I, Mark D, Vincent JH, *Investigation of performance of personal samplers in industrial workplaces*. 1988, Institute of Occupational Medicine: Edinburgh, UK.
183. Cherrie JW, Stelfox R, *Airborn dust levels generated during insulation with MMMF: a laboratory investigation*. 1990, Institute of Occupational Medicine: Edinburgh, UK.
184. Sahle W, Sallsten G, Thoren K. Characterization of airborne dust in a soft paper mill. *Ann Occup Hyg*. 1990;34:55-75.
185. Cherrie JW, Lynch G, Corfield MM, Cowie HA, Robertson A, *Effects of workers' behaviour on measurements made using pumped and diffusive techniques*. 1991, Institute of Occupational Medicine: Edinburgh, UK.
186. Ogden TL, Bartlett IW, Purnell CJ, Wells CJ, Armitage F, Wolfson H. Dust from cotton manufacture: changing from static to personal sampling. *Ann Occup Hyg*. 1993;37:271-85.

187. Cherrie JW, Gibson H, McIntosh C, Maclaren W, Lynch G. Exposure to fine airborn aramid fibrous dust amongst processors of Para-Aramid. *Ann. occup. Hyg.* 1995;39:403-426.
188. Cherrie JW. The effect of room size and general ventilation on the relationship between near and far-field concentrations. *Appl Occup Environ Hyg.* 1999;14:539-46.
189. Westerlund J, Bryngelsson IL, Löfstedt H, Eriksson K, Westberg H, Graff P. Occupational exposure to trichloramine and trihalomethanes: adverse health effects among personnel in habilitation and rehabilitation swimming pools. *J Occup Environ Hyg.* 2019;16:78-88.
190. Westerlund J, Graff P, Bryngelsson IL, Westberg H, Eriksson K, Lofstedt H. Occupational Exposure to Trichloramine and Trihalomethanes in Swedish Indoor Swimming Pools: Evaluation of Personal and Stationary Monitoring. *Ann Occup Hyg.* 2015;59:1074-84.
191. Seys SF, Feyen L, Keirsbilck S, Adams E, Dupont LJ, Nemery B. An outbreak of swimming-pool related respiratory symptoms: An elusive source of trichloramine in a municipal indoor swimming pool. *Int J Hyg Environ Health.* 2015;218:386-91.
192. Helenius I, Rytila P, Sarna S, et al. Effect of continuing or finishing high-level sports on airway inflammation, bronchial hyperresponsiveness, and asthma: a 5-year prospective follow-up study of 42 highly trained swimmers. *J Allergy Clin Immunol.* 2002;109:962-8.
193. Pijnenburg MW, Floor SE, Hop WC, De Jongste JC. Daily ambulatory exhaled nitric oxide measurements in asthma. *Pediatr Allergy Immunol.* 2006;17:189-93.
194. Mattes J, Storm van's Gravesande K, Moeller C, Moseler M, Brandis M, Kuehr J. Circadian variation of exhaled nitric oxide and urinary eosinophil protein X in asthmatic and healthy children. *Pediatr Res.* 2002;51:190-4.
195. Shah D. Healthy worker effect phenomenon. *Indian J Occup Environ Med.* 2009;13:77-9.