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Ph.D. Thesis

Control of particulate matter emissions from poultry and pig houses

Control de las emisiones de material particulado de granjas avícolas y porcinas

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To my family, agricultural engineers by tradition and
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A mi familia, ingenieros agrónomos por tradición e
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Abstract

Livestock housing, especially poultry and pigs, are major sources of particulate matter (PM). High ambient concentrations of PM can threaten human and animal health and welfare, as well as the environment. The best approach to reduce PM emissions from livestock houses seems to be to prevent it from being generated. Controlling PM at source not only reduces emissions but also improves inside air quality. Furthermore, data on particle morphology and chemical composition are essential to evaluate the likely exposure to PM on the one hand, and on the other hand, to develop control measures to reduce it. The research aim of this thesis was to acquire knowledge on where PM comes from in various livestock housing systems and to evaluate abatement techniques on reducing PM in relation with other pollutants.

This thesis is composed of four research studies and a review of the state-of-the-art of PM in and from livestock production systems, which is the background of this thesis. Firstly, known sources of PM were collected from different housing systems for poultry and pigs and experimentally aerosolized in a laboratory dust generator to collect fine and coarse PM samples. These samples were analyzed i) using scanning electron microscopy with X-ray microanalysis to develop comprehensive morphological and chemical source profiles; and ii) with optical particle counter to determine source particle-size distribution. Secondly, the developed source profiles from known sources as well as particle morphological characteristics extracted with digital image analysis software were used to investigate which particle characteristics were best to distinguish amongst specific sources. Thirdly, the previous information was used to quantify the contribution of the different sources to fine and coarse airborne on-farm PM emissions from livestock houses using two source apportionment models (expert systems and multivariate linear regression models). To do this, we sampled airborne on-farm fine and coarse PM at 14 different livestock locations for poultry (including broilers, laying hens in floor, and aviary system and turkey production) and pigs (including piglets, growing-finishing pigs, and dry-pregnant sow housings). Finally, the potential of air ionization for reducing PM concentrations and emissions from a pilot-scale broiler farm was evaluated and its effect on particle properties and other pollutants was assessed.

Our results indicated that the sources that contribute to PM are specific to livestock housing system and livestock species and that housing systems and livestock species determine particle diversity and heterogeneity. The laboratory dust generation process was successfully applied to develop comprehensive morphological and chemical source

profiles for feathers, feed, manure, hair, skin, wood shavings, and outside source. The developed source profiles and presented particle-size distributions are valuable to compare similarities and differences in particle types and will allow faster and more accurate qualitative and semi-quantitative estimations of source contributions in future studies. Our results also indicated that to apply source apportionment models in livestock houses, it is necessary to obtain not only particle chemical characteristics, but also morphological particle characteristics because they can make additional value to using only chemical characteristics when sources show distinctive and well defined individual particle morphology or differ in size. On average 69% of particles belonging to a mixture of sources from poultry and pig houses can be correctly assigned to their source based on the combinations of chemical and morphological characteristics in fine and coarse PM, and based on our results, it is the recommended approach to apportion all individual sources to PM in livestock houses. In the surveyed poultry houses, source contributions vary amongst poultry housing systems, but most particles originate from feathers (ranging from 4 to 43% in fine and from 6 to 35% in coarse PM) and from manure (ranging from 9 to 85% in fine and from 30 to 94% in coarse PM). In the surveyed pig houses, source contributions vary amongst pig housing systems, but most particles originate from manure (ranging from 70 to 98% in fine and from 41 to 94% in coarse PM). When expressed in mass, big particles from wood shavings and especially skin gain relative importance compared with number of particles. Finally, air ionization proved to effectively and significantly reduce total PM10 mass emission by 36% and PM2.5 mass emissions by 10% in broiler production, but it had no effect on airborne micro-organisms, odor or ammonia emissions. Overall, the studies presented in this thesis have provided new knowledge for better and more efficient designing of PM reduction measures at source and for predicting how different techniques will work.

Resumen

Los alojamientos ganaderos, especialmente avícolas y porcinos, son una fuente importante de material particulado ("particulate matter", PM). Las concentraciones elevadas de PM en el ambiente pueden afectar a la salud de las personas y animales, así como al medio ambiente. La mejor manera de reducir las emisiones de PM de los alojamientos ganaderos es evitar que éste se genere y así, controlando el PM en origen, no sólo se pueden reducir las emisiones, sino también mejorar la calidad del aire en el interior de los alojamientos ganaderos. Por otra parte, para evaluar la posible exposición al PM por un lado, y para desarrollar medidas para reducirlo, por otro, es necesario conocer la morfología y composición de las partículas. En consecuencia, el objetivo de esta tesis fue identificar y caracterizar el origen del PM en diferentes sistemas de alojamientos ganaderos y evaluar técnicas de reducción de dicho PM en relación con otros contaminantes.

La tesis está compuesta por cuatro trabajos de investigación y una revisión previa, sobre el estado de la cuestión del PM en los sistemas de producción ganaderos, que establece el marco del trabajo experimental. En primer lugar, se muestrearon fuentes conocidas de PM en alojamientos ganaderos que fueron aerosolizadas experimentalmente en un generador de polvo de laboratorio para recoger muestras de PM fino y grueso. Estas muestras fueron analizadas posteriormente mediante: i) microscopía electrónica de barrido con un espectrómetro de rayos X para obtener una caracterización morfológica y química detallada de las fuentes; ii) mediante un contador óptico de partículas para obtener la distribución por tamaños de cada fuente. En segundo lugar, se investigaron las características más adecuadas de las partículas para distinguir entre las distintas fuentes en base a la caracterización de las mismas anteriormente obtenida y a las características morfológicas obtenidas con análisis digital de imagen. En tercer lugar, se utilizó la información anterior para cuantificar la contribución de cada fuente al PM fino y grueso del aire de alojamientos ganaderos, mediante dos modelos de reparto de las contribuciones de PM (sistemas expertos y modelos de regresión lineal multivariante). Para ello, muestreamos PM suspendido en el aire de 14 alojamientos avícolas (incluyendo sistemas de producción para broilers, gallinas ponedoras en sistema tipo suelo y aviario, y pavos) y porcinos (incluyendo lechones, cerdos de cebo y cerdas secas y gestantes). Finalmente, se evaluó el potencial de la técnica de ionización del aire para reducir las concentraciones y emisiones de PM en una granja piloto de broilers y se valoró su efecto sobre las propiedades de las partículas y sobre otros contaminantes.

Los resultados indican que la diversidad y heterogeneidad de las partículas de las distintas fuentes de alojamientos ganaderos está determinada por las propias fuentes que proporcionan el PM, que son específicas de los sistemas de alojamiento ganaderos y de la especie animal. El proceso de generación experimental de partículas desarrollado fue adecuado para realizar una caracterización detallada de fuentes tales como plumas, pienso, estiércol, pelo, piel, viruta de madera y entorno exterior en las granjas. Esta caracterización junto con la distribución del tamaño de las partículas de cada fuente fue útil establecer similitudes y diferencias entre los tipos de partículas, lo que permitirá realizar estimaciones cualitativas o semi-cuantitativas más precisas y rápidas de las fuentes que contribuyen al PM en alojamientos ganaderos en trabajos futuros. Los resultados obtenidos indican que para aplicar modelos de reparto de las contribuciones de PM en alojamientos ganaderos, es necesario obtener no sólo las características químicas de las partículas sino también las características morfológicas de éstas porque pueden aportar un conocimiento adicional, cuando las partículas en cada fuente tienen una morfología individual bien definida y distintiva o difieren en su tamaño. Utilizando la combinación de las características químicas y morfológicas, se puede asignar correctamente a cada una de sus fuentes una media de 69% de las partículas procedentes de una mezcla de fuentes en alojamientos avícolas y porcinos en el PM fino y grueso. Según resultados obtenidos, este es el enfoque recomendado para repartir el PM generado en alojamientos ganaderos por las distintas fuentes individuales. En efecto, las contribuciones de las distintas fuentes varían con el sistema de alojamientos: en los alojamientos avícolas muestreados, la mayoría de las partículas se originan a partir de las plumas (rango entre 4 a 43% PM fino y entre 6 a 35% en el PM grueso) y de la gallinaza (rango entre 9 a 85% PM fino y entre 30 a 94% en el PM grueso); mientras que en los alojamientos porcinos, la mayoría de las partículas se originan a partir del estiércol (rango entre 70 a 89% PM fino y entre 41 a 94% en el PM grueso). Las partículas de viruta de madera y de piel animal adquieren mayor importancia relativa cuando se expresan estas contribuciones en masa de partículas. Finalmente, se demostró que la ionización del aire pudo reducir eficaz y significativamente la emisión total en masa de PM10 en un 36% y la de PM2.5 en un 10% en la producción de broilers, pero que no mostró ningún efecto en los microorganismos suspendidos, sobre los olores o sobre le emisión de amoniaco. En su conjunto, se puede concluir de manera genérica que los resultados presentados en esta tesis contribuyen a proporcionar unas herramientas básicas que permitirán diseñar unas medidas de reducción de PM en origen mejores y más eficientes y, paralelamente, a predecir su funcionamiento.

Resum

Els allotjaments ramaders, especialment avícoles i porcins, són una font important de material particulat ("particulate matter", PM). Les concentracions elevades de PM en l'ambient poden afectar a la salut de les persones i dels animals, així com al medi ambient. El millor mode de reduir les emissions de PM dels allotjaments ramaders és evitar que aquest es genere i així, controlant el PM en origen, no solament es poden reduir les emissions, sinó també millorar la qualitat de l'aire a l'interior dels allotjaments ramaders. Per tal d'avaluar la possible exposició al PM d'una banda, i per a desenvolupar mesures per a reduir-ho, per una altra, és necessari conèixer la morfologia i composició de les partícules. En conseqüència, l'objectiu d'aquesta tesi és identificar i caracteritzar l'origen del PM en diferents sistemes d'allotjaments ramaders i avaluar tècniques de reducció de d'un determinat PM en relació amb altres contaminants.

La tesi comprèn quatre treballs de recerca i una revisió prèvia, sobre l'estat de la qüestió del PM en els sistemes de producció ramaders, que estableix el marc d'aquesta tesi. En relació als treballs, en primer lloc, es mostrejaren fonts conegudes de PM en allotjaments ramaders que van ser aerosolitzades experimentalment en un generador de pols del laboratori per a arreplegar mostres de PM fi i gruix. Aquestes mostres van ser analitzades posteriorment mitjançant: i) microscòpia electrònica d'escombratge amb un espectròmetre de rajos X per a obtenir una caracterització morfològica i química detallada de les fonts; ii) mitjançant un comptador òptic de partícules per a obtenir la distribució per grandàries de cada font. En segon lloc, es van investigar les característiques més adequades de les partícules per a distingir entre les diferents fonts basant-se en la caracterització de les fonts anteriorment obtinguda i a les característiques morfològiques obtingudes amb anàlisi digital d'imatge. En tercer lloc, es va utilitzar la informació anterior per a quantificar la contribució de cada font al PM fi i gruix de l'aire d'allotjaments ramaders mitjançant dos models de repartiment de les contribucions de PM (sistemes experts i models de regressió lineal multivariant). Per a açò, mostrejem PM suspès en l'aire de 14 allotjaments avícoles (incloent sistemes de producció per a broilers, gallines ponedores en sistema tipus sol i aviari, i galls dindis) i porcins (incloent garrins, porcs d'esquer i truges seques i gestants). Finalment, es va avaluar el potencial de la tècnica de ionització de l'aire per a reduir les concentracions i emissions de PM en una granja pilot de broilers i es va valorar el seu efecte sobre les propietats de les partícules i sobre altres contaminants.

Els resultats indiquen que la diversitat i heterogeneïtat de les partícules de les diferents fonts d'allotjaments ramaders està determinada per les pròpies fonts que contribueixen al PM, que són específiques dels sistemes d'allotjament ramaders i de l'espècie animal. El procés de generació experimental de pols desenvolupat va ser adequat per a realitzar una caracterització detallada de fonts tals com a plomes, pinso, fem, pèl, pell, encenall de fusta i de l'entorn exterior de les granges. Aquesta caracterització juntament amb la distribució de la grandària de les partícules de cada font és útil per a comparar similituds i diferències entre els tipus de partícules i permetrà realitzar estimacions qualitatives o semi-quantitatives més precises i ràpides de les fonts que contribueixen al PM en allotjaments ramaders en treballs futurs. Els resultats obtinguts indiquen que per a aplicar models de repartiment de les contribucions de PM en allotjaments ramaders, és necessari obtenir no sols les característiques químiques de les partícules si no també les característiques morfològiques d'aquestes perquè poden aportar un coneixement addicional quan les partícules en cada font tenen una morfologia individual ben definida i distintiva o difereixen en la seua grandària. Utilitzant la combinació de les característiques químiques i morfològiques de les partícules, es pot assignar correctament a cadascuna de les seues fonts, una mitjana de 69% de les partícules procedents d'una mescla de fonts d'allotjaments avícoles y porcins en el PM fi i gruix. Segons resultats obtinguts, aquest és l'enfocament recomanat per a repartir el PM generat en allotjaments ramaders per les fonts individuals. En efecte, les contribucions de les diferents fonts varien amb el sistema d'allotjament. Així, en els allotjaments avícoles mostrejats, la majoria de les partícules s'originen de les plomes (rang entre 4 a 43% PM fi i entre 6 a 35% en el PM gruixut) i del fem (rang entre 9 a 85% PM fi i entre 30 a 94% en el PM gruixut). D'altra banda, en els allotjaments porcins, la majoria de les partícules s'originen del fem (rang entre 70 a 89% PM fi i entre 41 a 94% en el PM gruixut). Les partícules d'encenall de fusta i de pell animal adquireixen major importància relativa quan s'expressen aquestes contribucions en massa de partícules. La ionització de l'aire pot reduir eficaçment i significativament l'emissió total en massa de PM10 en un 36% i la de PM2.5 en un 10% per a broilers, però no mostra cap efecte en els microorganismes suspesos, sobre olors o sobre amoníac. En el seu conjunt, es pot concloure de manera genèrica que els resultats presentats en aquesta tesi contribueixen a proporcionar unes eines bàsiques que permetran dissenyar unes mesures de reducció de PM en origen millors i més eficients i, paral·lelament, a predir el seu funcionament.

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Chapter 1

General introduction and outline

1.1.   Justification: the problem

Atmospheric pollution can be defined as a change in the substances present in ambient air which are likely to be harmful for humans and other living organisms and/or for the natural environment. In this sense, airborne particulate matter (PM) is an important pollutant because it can cause serious health problems such as respiratory and cardiovascular disease and increased mortality (Dockery *et al.*, 1993; Pope *et al.*, 2002) and it can cause reduced visibility, vegetation stress, and ecosystems alteration (Grantz *et al.*, 2003) as well as it can affect the Earth's radiative balance (IPCC, 2001).

Besides traffic and industrial activities, livestock housing is a major source of PM. Generated PM inside livestock houses is emitted to the external environment through the ventilation exhausts. Therefore, there is growing concern that in certain regions where background PM concentrations due to other sources are already high, PM emitted from livestock houses can cause exceedance of the limits established by the European air quality regulations. Limits are set for two types of PM, depending on its size: PM10 and PM2.5 (fine PM). The annual average limit for PM10 is set to 40 μg m^{-3} (Directive 1999/30/EC). The annual average limit for PM2.5 is set to 25 μg m^{-3} to be met by 2015 (Directive 2008/50/EC). The PM10 fraction can be inhaled and accumulated in the upper respiratory airways. This fraction includes the smaller PM2.5 fraction (fine PM) which can penetrate deeper in the respiratory airways and can reach the alveoli in the lungs. Consequently, size is one of the most important properties related to PM because it is critical to its health effects. Particle size also has a direct influence on PM atmospheric behavior, affecting its atmospheric lifetime, transport, and fate. Therefore, particle size is also critical to its environmental effects.

With the diversity of livestock production systems, PM can originate from a wide variety of sources and this results in PM from livestock houses being very heterogeneous in composition and morphology. In livestock houses, feed, manure, bedding, and animal's skin, feathers, and hair have been identified as the main sources of PM (Aarnink *et al.*, 1999; Donham *et al.*, 1986; Feddes *et al.*, 1992; Heber *et al.*, 1988; Qi *et al.*, 1992). The generation of PM, its suspension in the air and its release outside livestock houses depends on kind of housing and feeding, animal type, and environmental

factors related with climatic conditions (Takai *et al.*, 1998). Despite its heterogeneity, PM from livestock houses is unique not only because it has a high organic content (above 90%) and it comprises many sizes, shapes, densities, and chemical compositions; but also because it can act as a carrier of other substances which are highly generated and available inside livestock houses. Particles can adsorb odorous compounds and gases (Cai *et al.*, 2006) and they can also contain biologically active micro-organisms (Lee *et al.*, 2006). Exposure to PM from livestock houses and to its compounds can pose serious damage to the respiratory health of farmers and people living in the vicinity of farms, and cause negative environmental effects (Andersen *et al.*, 2004; Wathes *et al.*, 2004; Whyte, 2002).

To date we still do not know the extent of the potential health hazards of PM from livestock housing systems to the population, as there is little information on specific particle characteristics such as morphology and chemical and microbiological composition. Information on mass concentrations are available, but mass-only measurements are not enough to fully understand health and environmental effects, as these may be related to PM characteristics other than mass. Moreover, existing data on particle morphology and composition are not sufficient to solve PM related problems, thus there is need for this information to know the nature of the problem and to evaluate the likely exposure to PM on the one hand, and on the other hand, to develop control measures to reduce this pollutant.

Available abatement techniques are being tested and evaluated to reduce PM in and from livestock houses (Aarnink *et al.*, 2009; Pedersen *et al.*, 2000; Takai and Pedersen, 2000). Although there is a wide range of available options, some of them fail to work in practice when they are challenged under farm conditions, with different animal types and farm situations. The best approach to reduce PM in and from livestock houses seems to be to prevent it from being generated. Controlling PM at source not only reduces PM emissions but also improves inside air quality. To this end, it is essential to identify exactly where PM comes from in the diversity of housing systems and livestock categories. In this way it will be possible to tackle PM pollution at source in livestock houses and to optimize the efficiency and technical and economical viability of some of the most promising and available reduction techniques. Only by knowing the problem and with a

complete characterization of PM from livestock houses, efficient and more effective reduction measures can be developed and implemented to comply with air quality regulations and contribute to reduce PM pollution from livestock houses.

1.2. Objectives and outline of the thesis

The general research aim of this thesis is to acquire knowledge on where PM comes from in various livestock housing systems and to evaluate abatement techniques on reducing PM in relation with other pollutants. These steps are essential to identify effective systems, techniques and combinations of both to control this pollutant at source. With this aim, the following specific objectives were defined:

1. To review the state-of-the-art of PM in and from livestock production systems and identify future research priorities to characterize and reduce PM from livestock houses.

2. To morphologically and chemically characterize several known sources of PM collected from housing systems for poultry and pigs in different size fractions and to develop detailed source profiles.

3. To identify the most effective particle characteristics to distinguish amongst sources of PM in different size fractions in poultry and pig housing systems.

4. To quantify the individual contribution of sources to airborne on-farm PM emissions in different size fractions in poultry and pig housing systems.

5. To assess the effectiveness of a PM reduction technique in a case-study and fully evaluate its effects on PM concentration, emission, particle properties, and its effect on other pollutants.

This thesis is divided in seven chapters. A review of the state-of-the-art of PM in and from livestock production systems was the background of this thesis and is presented in Chapter 2, in which the key research needs related

to the control of PM in livestock houses are described and discussed. As one of the main areas of knowledge deficiency identified was PM characterization and source apportionment, these aspects were dealt with and emphasized in Chapters 3, 4 and 5. Chapter 3 provides a complete and detailed analysis of known sources of PM which can potentially contribute to PM in poultry and pig houses and a qualitative description of source morphology and elemental composition can be found herein. This chapter provides the basis for Chapters 4 and 5. Chapter 4 describes which particle characteristics are best to discriminate between each individual source in each livestock species studied (poultry and pigs) and identifies the most efficient methods to distinguish amongst them. The results obtained in Chapter 4 were applied in Chapter 5, in which the contribution of each individual source to airborne on-farm PM is quantified at 14 different livestock locations for poultry (including broilers, laying hens in floor and aviary system and turkey production) and pigs (including piglets, growing-finishing pigs, and dry-pregnant sow housings). Chapter 6 is a case-study where the potential of air ionization for reducing PM concentrations and emissions from a pilot-scale broiler farm is evaluated and its effect on particle size and size distribution, as well as on ammonia, odor, and micro-organisms are assessed.

The main results from this thesis are discussed in a broader context and their implications for future research on how to control PM in and from livestock houses are described in Chapter 7. In this chapter, the main conclusions of this thesis are also drawn.

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Chapter 2

Airborne particulate matter from livestock production systems: A review of an air pollution problem

Airborne particulate matter from livestock production systems:

A review of an air pollution problem

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Abstract. Livestock housing is an important source of emissions of particulate matter (PM). High concentrations of PM can threaten the environment, as well as the health and welfare of humans and animals. Particulate matter in livestock houses is mainly coarse, primary in origin, and organic; it can adsorb and contain gases, odorous compounds, and micro-organisms, which can enhance its biological effect. Levels of PM in livestock houses are high, influenced by kind of housing and feeding, animal type, and environmental factors. Improved knowledge on particle morphology, primarily size, composition, levels, and the factors influencing these can be useful to identify and quantify sources of PM more accurately, to evaluate their effects, and to propose adequate abatement strategies in livestock houses. This paper reviews the state-of-the-art of PM in and from livestock production systems. Future research to characterize and control PM in livestock houses is discussed.

Glossary of definitions and abbreviations

Term	Definition	Acronym
Aerosol	Same as particulate matter	
Aerodynamic diameter	Diameter of a sphere particle with a density of 1 g cm^{-3} that would have the same settling velocity as the particle in question	AED
Aluminum	Chemical element Al	Al
Ammonia	Chemical compound NH_3	NH_3
Ammonium	NH_4^+ ion	NH_4^+
Ammonium bisulfate	Chemical compound formed by reaction of ammonia and sulfuric acid	NH_4HSO_4
Ammonium chloride	Chemical compound formed by reaction of ammonia and hydrochloric acid	NH_4Cl
Ammonium nitrate	Chemical compound formed by reaction of ammonia and nitric acid	NH_4NO_3
Ammonium sulfate	Chemical compound formed by reaction of ammonia and sulfuric acid	$(NH_4)_2SO_4$
Beta-glucans	Polysaccharides found in the outer layer of cereal grains, the cell wall of baker's yeast, some fungi, and some mushrooms	
Bio-aerosols	Particles of biological origin and/or activity	
Brownian motion	Random movement of particles suspended in a liquid or gas medium	
Calcium	Chemical element Ca	Ca
Chloride	Chemical element Cl	Cl
Coagulation	Aggregation of small particles to form larger particles, increasing size and thus easier to remove from an airstream	
Coarse PM	Particles bigger than 2.5 µm and smaller than 10 µm in diameter	PM10-2.5
Condensation	Change of shape and size of particles when gas or vapor molecules emitted at high temperatures condense in contact with cooler air, on the surface of existing particles	
Crustal material	Earth material related to soil	
Diffusion	Primary means of transportation and deposition of small airborne particles and gas molecules as a result of a concentration gradient at low air velocities	
Dry matter	Chemical determination of mass of something with all its constituents excluding water	DM
Dust	Solid particles (settled or airborne) formed by mechanical fracture of a parental material, which can sediment under gravity forces, with diameters up to 500 or 1000 µm	
Elemental carbon	Soot, black carbon, or light absorbing carbon	
Endotoxins	Lipopolysaccharide complexes originating from the membrane of Gram negative bacteria	

Endotoxin unit	Endotoxin activity of about 0.1 ng of Reference Endotoxin Standard	EU
European Standardization Committee	Platform for the development of European Standards	CEN
Fine PM	Particles smaller than 2.5 µm in diameter	PM2.5
Gas-to-particle conversion	Chemical reactions between gases causing particle formation in the atmosphere	
Geometric standard deviation	Geometric standard deviation of a particle-size log-normal distribution	σ_g
Gravitational settling	Deposition of airborne particles by gravitational forces	
Hydrochloric acid or hydrogen chloride	Chemical compound HCl, which can react with ammonia to form secondary ammonium chloride	HCl
Hydrogen sulfide	Odorous chemical compound H_2S	H_2S
Inhalable PM	Mass fraction of total airborne particles which can be inhaled through nose and mouth	
International Standards Organization	International Organization for Standardization for business, government, and society	ISO
IPPC	Integrated Prevention Pollution and Control	IPPC
Magnesium	Chemical element Mg	Mg
Mass median diameter	Mass median aerodynamic diameter of a particle used to describe particle-size log-normal distributions	d_{50}
Micrometer	Unit of length, 10^{-6} of a meter	µm
Nitrates	Chemical compound with NO_3^- group, or NO_3^- ion	NO_3^-
Nitric acid	Chemical compound HNO_3	HNO_3
Nitrogen	Chemical element N	N
Nitrogen oxides	Nitrogen oxides typically emitted from combustion processes. They can react with water to form nitric acid which can react with ammonia to form secondary ammonium nitrate particles. They are precursor gases	NO_x
Nonanal	Odorous aldehyde chemical compound	$C_9H_{18}O$
Nucleation	Particle formation and growth from gas or vapor phase, to liquid (droplet) or solid phase, around an existing particle or nucleus	
Octanal	Odorous aldehyde chemical compound	$CH_3(CH_2)_6CHO$
Particle-size distribution	Relative proportion of particle mass (volume), surface, and number in a certain size range	PSD
Particulate matter	Fine solid or liquid particles suspended in a gaseous medium (same as aerosol)	PM
Phosphorus	Chemical element P	P
PM2.5	Particulate matter which passes through a size-selective inlet with a 50 % efficiency cut-off at 2.5	

	μm aerodynamic diameter	
PM4	Particulate matter which passes through a size-selective inlet with a 50 % efficiency cut-off at 4 μm aerodynamic diameter	
PM5	Fine particulate matter with 50% cut-off diameter of 5 μm, according to the Convention of Johannisburg	
PM10	Particulate matter which passes through a size-selective inlet with a 50 % efficiency cut-off at 10 μm aerodynamic diameter	
Potassium	Chemical element K	K
Precursor	Any chemical (usually gaseous) which can contribute to gas-to-particle conversion	
Primary PM	Particles which are directly emitted to the atmosphere	
RAINS model	Regional Air Pollution Information and Simulation Model under the International Institute for Applied Systems Analysis (IIASA)	
Respirable PM	Mass fraction of inhaled particles which can go beyond the larynx and penetrate into the unciliated respiratory system	
Secondary PM	Particles which are formed in the atmosphere by gas-to-particle conversions	
SEM-EDX	Scanning electron microscopy with X-ray single particle analysis	
Silicon	Chemical element Si	Si
Sodium	Chemical element Na	Na
Submicron particles	Particles smaller than 1 μm in diameter	PM1
Sulfates	Chemical compounds with SO_4^{2-} group, or SO_4^{2-} ion	SO_4^{2-}
Sulfur	Chemical element S	S
Sulfur dioxide	Chemical compound SO_2, which can react with hydrogen to form sulfuric acid. It is a precursor gas	SO_2
Sulfuric acid	Chemical compound H_2SO_4	H_2SO_4
Thoracic PM	Mass fraction of inhaled particles which can penetrate into the larynx	
Total suspended particles	All airborne particles suspended in ambient calm air or dispersed in an air flow. It refers to particles smaller than 30 to 100 μm in diameter depending on the sampling conditions	TSP
Trace element	Chemical element found in very small amounts in PM (e.g. lead or mercury)	
Volatile organic compounds	Molecules containing carbon, with high enough vapor pressures under normal conditions to vaporize and enter the atmosphere (e.g. light hydrocarbons, ketones, and aldehydes)	VOCs
Ultra-fine particles	Particles which range from 50 to 200 nm in diameter	

2.1.   Introduction

Traditionally, particulate matter (PM) from livestock houses has been regarded as an indoor pollutant causing detrimental effects on animal performance and efficiency (Al Homidan and Robertson, 2003; Donham and Leininger, 1984), and on the health and welfare of farmers (Andersen *et al.*, 2004; Donham *et al.*, 1984). Inhaled particles can penetrate in the deeper respiratory airways, compromising animal's and human's respiratory health, contributing to increased occurrence of chronic cough and/or phlegm, chronic bronchitis, allergic reactions and asthma-like symptoms amongst livestock farmers (Donham, 2000; Radon *et al.*, 2001; Zuskin *et al.*, 1995).

Research has revealed that PM aspects are not only important due to air quality issues inside, but also outside livestock houses (Pope *et al.*, 2002). Through the ventilation exhausts, pollutants generated in livestock houses are released to the outside environment (Phillips *et al.*, 1998). Agricultural activities in general and livestock production in particular can emit considerable amounts of PM to the atmosphere (Takai *et al.*, 1998). Although the share of emissions from livestock production in Europe only represents 8% of total PM10 emissions, and 4% of total primary PM2.5 emissions, agricultural sources are expected to gain relative importance because emissions from other sources will decline in the future according to current legislation baseline projections (Grimm, 2007; Klimont *et al.*, 2007). In The Netherlands, for instance, the contribution of agriculture to PM emissions is estimated to be approximately 25% (Chardon and van der Hoek, 2002). Inside livestock production, intensive poultry and pig houses are the main sources of PM emissions contributing to about 50% (poultry), and 30% (pigs) of total PM emissions from agriculture in Europe (EMEP-CORINAIR, 2007).

Outside livestock houses, besides compromising health and welfare of humans and animals, emitted PM can also threaten the environment (plants and other organisms) causing vegetation stress and ecosystem alteration (Grantz *et al.*, 2003). High concentrations of PM are, furthermore, relevant to climate change issues, such as cloud formation, radiative forcing, and contribute to atmospheric visibility impairment (IPCC, 2005). Intensification of livestock production in localized areas can contribute to

increasing environmental PM concentrations, and this can become a major issue, especially in areas where background PM concentrations due to other sources such as traffic or industry are already high.

High concentrations of environmental PM can be harmful itself and it can carry gases and odors, micro-organisms and their components, and other bioactive components (Bakutis *et al.*, 2004; Cai *et al.*, 2006; Seedorf, 2004) which can be transported away from the source causing respiratory affections to people living in the vicinity of the farms, as well (Radon *et al.*, 2007). Ammonia (NH_3) emitted from livestock houses is also a main precursor for forming secondary inorganic particles in the atmosphere (Erisman and Schaap, 2004).

These are the reasons why PM issues have found entrance into national and international regulations and strategies of air pollution and control (Integrated Prevention Pollution and Control, IPPC Directive 1996/61/EC, Council Directive 1999/30/EC, Directive 1996/62/EC and Directive 2008/50/EC), and there is growing pressure to evaluate and control PM emissions from main sources including intensively housed livestock.

To protect the environment and to ensure health and welfare of humans and animals in and around livestock houses, the concentrations and emissions of PM within such buildings must be known (Hinz and Linke, 1998b). It is necessary on the one hand, to obtain data on two fundamental particle properties: particle morphology, primarily size, and composition, to understand how PM is formed in livestock houses. This information can be useful to specifically identify and quantify sources of PM (Casuccio *et al.*, 2004). Little is known, however, about these two fundamental particle properties. To better understand the processes and factors which contribute to PM being emitted from livestock houses, on the other hand, it is also necessary to quantify the amount of PM which is emitted. There is still lack, however, of available data which offers reliable and comparable information and emission estimates which can contribute to fulfill this gap of knowledge.

The aim of this work is to give an overview of the major problems associated with PM in livestock production systems. This paper critically reviews the state-of-the-art of PM in and from livestock production systems, with emphasis on particle morphology and composition. Current

investigations of PM sources, levels (concentrations and emissions), the factors influencing these levels, particle size, and composition, are reviewed. Abatement strategies to reduce PM and its adverse effects are evaluated. Future research and strategies to characterize and control PM from livestock houses are discussed.

2.2. Particulate matter description and classification

Particulate matter is not a single pollutant, but a mixture of many types of pollutants. A particle can be defined as a small, discrete object, and PM includes materials with particle-like properties. The term PM is often used for air quality applications, to refer to fine solid or liquid particles suspended in a gaseous medium. This definition is also true for the term aerosol, although this term is more commonly used in atmospheric science. The term dust is used to refer only to solid particles of matter formed by mechanical fracture (i.e. crushing) of a parental material, which sediment under gravity forces (Zhang, 2004). The term PM can therefore be defined as a complex mixture of suspended particles with different physical, chemical and biological characteristics, which determine both its behavior, as well as its environmental and health effects (EPA, 2004).

The heterogeneous nature of PM comprises particles of different nature, shape, size, density, and chemical composition. This heterogeneity also applies to PM from livestock houses. Figure 2.1 shows, as an example, flake, oval, and crystalline shaped particles with sizes ranging from few nanometers (nm) to tens of micrometers (µm) in diameter, of PM from a rabbit house (Cambra-López and Torres, 2008). Particulate matter in livestock houses differs from other types of particles for three reasons: its concentrations are generally 10 to 100 times higher than in other indoor environments, it is an odor and gas carrier, and it is biologically active, generally containing a great variety of bacteria and micro-organisms (Zhang, 2004).

The standard scientific term to describe the behavior of particles in the atmosphere or in the human respiratory tract, is the aerodynamic equivalent diameter (AED). The AED groups three particle properties in a single parameter: size, shape, and density. It is defined as the diameter of a sphere particle with a density of 1 g cm^{-3} that would have the same settling velocity

as the particle in question (Baron and Willeke, 1993). This diameter is a useful measurable index for irregular shaped particles, because particles with the same AED presumably perform alike when suspended in the air.

Figure 2.1. Scanning electron micrograph (1500x) of heterogeneous PM from a rabbit house, collected on glass fiber filter. Adapted from Cambra-López and Torres (2008).

Particle-size distribution (PSD) is another useful term given the heterogeneity of AED in a mixture of particles. The PSD depends on the origin of particles, but a broad range of particle sizes can apply to the same origin. A PSD is used to quantify the proportion of PM sample whose AED is within a certain range, expressed as a fractional or cumulative frequency distribution. Some PSD follow a normal curve if you plot particle mass or number vs. particle size on a logarithmic scale. The mass median diameter (d_{50}) and the geometric standard deviation (σ_g) of a log-normal frequency distribution are used, amongst others, to describe more accurately PSD by statistical means (Baron and Willeke, 1993).

Different conventions and approaches are used to classify PM (EN 481, ISO 7708 and US EPA). Approaches include occupational health sizes, sampling cut-off sizes, and modes of distribution. It is complicated to classify PM into a single category or type. Therefore it is important to be clear on which classification has been used, and to provide definitions and penetration curves, to facilitate comparison of results.

Occupational health sizes are defined by the International Standards Organization (ISO), in ISO 7708 (ISO, 1995), and the European Standardization Committee (CEN), in EN 481 (EN, 1993). Occupational

health sizes are based on the behavior of particles in the human respiratory tract, and are derived from the depth of entrance into it. Human health-related sizes according to these conventions are: inhalable (particles which can be inhaled through the nose and mouth), thoracic (particles inhaled which can penetrate into the larynx), and respirable (particles which can go beyond the larynx and penetrate into the unciliated respiratory system) (EN, 1993). The ISO 7708 (ISO, 1995) further defines a second respirable convention, for children, and sick or infirm: the high risk respirable.

Concerning ambient air, the US EPA Code of Federal Regulations (US EPA, 2001a and 2001b), and Council Directive 1999/30/EC defined PM10 and PM2.5. There is a tendency to relate outside air quality to PM10 and PM2.5, instead of to occupational health size fractions. The PM10 and PM2.5 fractions are defined as the sampling cut-off diameter of particle separators that the mass of total suspended particles (TSP) have to pass, for a separation or sampling efficiency of 50%. This varies with the type of sampler and sampling efficiency.

Sampling criteria and particle penetration curves are given in the conventions simulating the different breathable particle fractions. Penetration curves define the sampling performance of a sampler in terms of particle mass fraction collected in a region, for particles up to 100 µm. With the shape of these curves, occupational health size fractions can be compared with US EPA PM10 and PM2.5 fractions (Figure 2.2). Therefore, PM10 is comparable to the thoracic fraction, although with differences in the range of particle diameters (thoracic considers up to 40 µm in diameter, compared with 15 µm for PM10). The PM2.5 fraction can be considered equivalent to the high risk respirable defined by the ISO 7708 (ISO, 1995). The respirable fraction described in EN 481 (EN, 1993) and ISO 7708 (ISO, 1995), however, is comparable to PM4, where 50% of all particles with AED of 4 µm are respirable, while 50% will be separated in the upper parts. The PM2.5 instead of PM4 is commonly used in outside air quality nomenclature, because PM2.5 is the fraction which contains fine and ultra-fine particles, with greater risks of adverse health effects.

Based on PSD and formation mechanisms, PM can also be classified in modes: nucleation, accumulation, and coarse mode. Modes are defined

primarily by formation mechanisms, but also differ in sources, composition, transport, fate, and size (EPA, 2004). Following the modal classification, PM is further classified into fine and coarse particles by size, and as primary and secondary by source or origin. Figure 2.3 illustrates these three PM classifications: by modes of distribution (nucleation, accumulation and coarse); by size (fine and coarse); and by origin (primary and secondary).
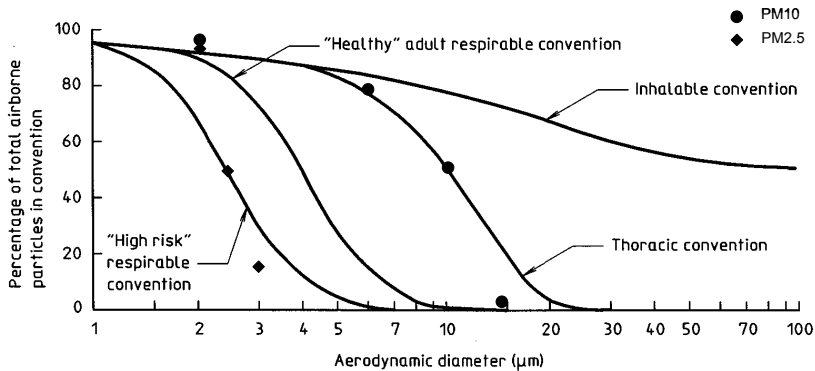
Figure 2.2. Inhalable, thoracic, respirable, and high risk respirable penetration curves defined in ISO 7708 (ISO, 1995) as percentage of total airborne particles, and PM10 and PM2.5 US EPA (2001a and 2001b) definitions. Adapted from ISO (1995).

As shown in Figure 2.3, nucleation mode is the smallest group in size, with particles with the smallest diameters, mainly directly emitted from the source, with short atmospheric lifetimes and subject to Brownian motion or diffusion. Particles in the nucleation mode can be formed by nucleation of gases or by condensation. These particles can easily coagulate to larger ones. The second mode, accumulation mode, also includes condensation particles. Accumulation mode particles have bigger d_{50} and longer atmospheric lifetimes than the nucleation mode particles, because these particles are too large to be subject to Brownian motion and too small to settle from air rapidly. Accumulation mode particles are usually eliminated from the atmosphere by washout, dry or wet deposition (AQEG, 2005). Dry deposition to plant and soil can occur by gravitational settling or inertial impaction of particles. Wet deposition can occur by precipitation of particles as rain or snow.
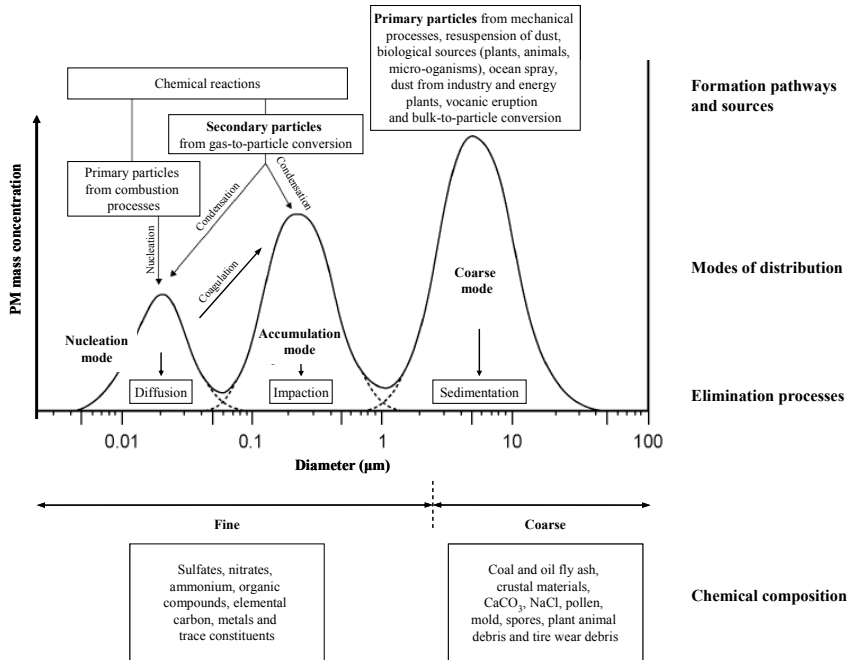
Figure 2.3. Schematic diagram of particle classification, size distribution, formation and elimination processes, modes of distribution and composition. Adapted from Seinfield and Pandis (1998) and EPA (2004).

The third mode, coarse mode, includes mechanically generated particles. Seinfeld and Pandis (1998) reported that particles in this mode have a minor health and environmental significance because they rapidly settle or sedimentate by gravitational forces. Particles in the coarse mode, however, can travel long distances depending on air speed. Many studies have established a relationship between coarse particles, and respiratory and cardiovascular disease (Brunekreef and Forsberg, 2005). The coarse fraction is, therefore, generally used as the main indicator of human exposure to PM, and adverse health effects are consistently related to physical exposure to coarse PM. Current European policies and PM standards aim precisely at this fraction of PM (e.g. Directive 1999/30/EC).

With regard to fine and coarse particles, the recommended cut-point between fine and coarse particles is generally 2.5 μm. The size separation between accumulation and coarse modes, being at approximately 1 μm, does not coincide with the 2.5 μm threshold to distinguish between fine and

coarse particles (Figure 2.3). This points out that some of the mechanically generated particles can fall into the PM2.5 size range. A more detailed classification also identifies ultra-fine particles as those smaller than 200 nm in diameter according to the Air Quality Directive (EFCA, 2009).

Primary and secondary PM can also be differentiated according to its origin, but also by the chemical composition of particles which varies with its origin, the same as its size (Almeida, 2006). On the one hand, primary particles are mainly coarse, emitted directly to the atmosphere, and are usually mechanically generated. Primary PM is mainly crustal, and rich in aluminum (Al) and silicon (Si), sodium (Na) and chloride (Cl) (Mazzei *et al.*, 2008). Primary PM includes particles of biological origin, rich in carbon (C), hydrogen (H), oxygen (O), nitrogen (N), phosphorus (P), and sulfur (S). These can include micro-organisms and their components, toxins, pollen, spores, and plant and animal debris. Particulate matter from housed livestock contains a much greater proportion of particles of biological origin and/or activity, usually referred to as bio-aerosols, compared with urban or industrial PM (Cox and Wathes, 1995).

On the other hand, secondary particles are fine, falling normally in the PM2.5 size range, and are generated from chemical reactions between gases and particles in the atmosphere. Some particles in the PM2.5 size range, however, can also be primary in origin, like particles emitted from combustion processes, rich in elemental carbon. Secondary PM is rich in sulfates (SO_4^{2-}), nitrates (NO_3^-), and ammonium (NH_4^+). Gas-to-particle conversion processes can occur to form secondary inorganic particles, in the presence of certain precursor gases such as NH_3, nitrogen oxides (NO_x), sulfur dioxide (SO_2), and volatile organic compounds (VOCs).

2.3.   To what extent is PM an air pollution problem?

Considering particles can come from a wide range of sources of origins, health and environmental hazards can vary considerably.

2.3.1.   Health hazards

Health hazards of PM can be separated between those related to inside (concentrations), and outside (emissions) livestock houses. The most relevant health hazards of PM inside livestock houses are related to

respiratory diseases (Andersen *et al.*, 2004). Increasing environmental PM concentrations outside, and exposure to PM in ambient air, is related to oxidative stress, respiratory diseases, and increased mortality. Recently reviewed epidemiological studies and health risks of PM described problems related to heart and lung disorders, and PM causing early deaths, especially in the elderly, children and ill persons (Buringh and Opperhuizen, 2002). Particles inside and outside livestock houses, moreover, might be a nuisance (e.g. caused by odorants adsorbed by airborne particles) (Wathes *et al.*, 2004).

There are three ways in which PM might affect health: by irritation of the respiratory tract and reduction of immune resistance to respiratory diseases by PM inhalation, by irritation of the respiratory tract by certain compounds present in PM, and by inhalation of pathogenic and non-pathogenic micro-organisms carried by PM (Harry, 1978).

The first way is related to PM itself. Physical properties of particles can cause harm when inhaled. A close relation between PM air pollution, respiratory and cardiovascular disease, and mortality, has been identified in the long term (Dockery *et al.*, 1993; Pope *et al.*, 2002), as well as in the short term (Ballester *et al.*, 2002; Hoek *et al.*, 2000). As livestock farmers are exposed to much higher PM concentrations inside livestock houses than in the outside air, the prevalence of respiratory diseases in livestock farmers is a lot higher than in other occupations (Bongers *et al.*, 1987; Donham *et al.*, 1984). Animal's respiratory health may also be compromised by PM (Al Homidan and Robertson, 2003; Donham and Leininger, 1984).

The first way in which PM can affect health is also related to the second and third way. Effects of PM on the respiratory systems of humans and animals may be aggravated by compounds present in PM. These compounds do not only affect human respiratory health, but also animal welfare and productivity through induced disease manifestation and increased mortality (Donham, 1991). Particles from livestock houses can contain a great number of substances such as heavy metals, VOCs, NO_3^-, and SO_4^{2-} which can be found adhered to the surface of particles (Martin *et al.*, 2008; Schneider *et al.*, 2001). Particulate matter can also adsorb irritant gases, especially NH_3 (Lee and Zhang, 2006; Takai *et al.*, 2002), and odorous

compounds (Das *et al.*, 2004; Razote *et al.*, 2004). Bioactive components can also be found attached to PM such as endotoxins, antibiotics, allergens, dust mites, and beta-glucans. Attached to fine PM, these compounds are claimed to increase the potential health hazard of PM if they have access to the deeper respiratory airways, enhancing the biological effect of PM (Donham and Leininger, 1984).

Particles can carry NH_3 molecules for a long time and can adsorb large amounts of NH_3 (up to 7 µg NH_3 per mg of respirable PM) (Takai *et al.*, 2002). The relative amount of NH_3 adsorbed on particles in livestock houses, mainly in PM2.5, can add up to 24% of total NH_3 in the gas phase (Reynolds *et al.*, 1998). This percentage, calculated using impingers with prefilters to measure gaseous and particulate NH_3, shows a relatively high proportion of NH_3 adsorbed on particles, compared with the gas phase. However, this is more likely to happen at low levels of ammonia as those reported in this study (NH_3 airborne concentrations below 7 ppm) than at higher ones, as those more commonly found in livestock houses. This needs to be looked at more carefully because of its implications when measuring NH_3 concentrations with other measuring techniques which do not account for NH_3 adsorbed on particles.

Odorants can be adsorbed and concentrated in PM, and consequently perceived as a more intense odor than odorants in the gas phase (Bottcher, 2001). More than 50 compounds bound to PM from a pig farm, belonging to different chemical classes, mainly alkanes, alcohols, aldehydes, ketones, acids, amines and nitrogen heterocycles, sulfides and thiols, aromatics, and furans have been identified (Cai *et al.*, 2006). The relative abundance of such compounds has shown to be higher in the smaller particles (Cai *et al.*, 2006). Significantly higher amounts of hydrogen sulfide (H_2S), octanal ($CH_3(CH_2)_6CHO$), and nonanal ($C_9H_{18}O$), were found in particles from 5 to 20 µm in diameter, than in larger particles in the range from 20 to 75 µm, in PM collected from different farms (Das *et al.*, 2004). Most of these compounds have already been identified as responsible for odor in livestock houses (O'Neill and Phillips, 1992; Schiffman *et al.*, 2001).

Most of the techniques, however, for measuring odors such as dynamic olfactometry, require dust filtering. If air samples are filtered, as in most

odor sampling devices, no odor from PM can be measured, and odor emissions might be underestimated in some cases. To establish relationships between odor and PM, the fraction of odor intensity related to particle-borne odors has to be known. It is difficult to measure this fraction because of filtering of air, and therefore, this relationship has not been accurately documented until now.

The third way in which PM might affect health, is related to bio-aerosols. Although the role of PM in airborne transmission of micro-organisms is not fully understood, PM is recognized as a vector for many micro-organisms. This was confirmed by the recovery of bacteria, fungi, as well as endotoxins from PM (Andersson *et al.*, 1999; Curtis *et al.*, 1975; Martin *et al.*, 1996). Bacteria mostly recovered from air in livestock houses are Gram positive bacteria with *Staphylococcus* and *Streptococcus* being predominant (Matkovic *et al.*, 2007). Some of these bacteria species have been recognized as to be responsible for many infections in human (Degener *et al.*, 1994; Gunn and Davis, 1988; Hedin and Widerstrom, 1998; Razonable *et al.*, 2001).

Although airborne Gram negative bacteria are found in relatively low percentage in livestock houses (less than 10%) compared with Gram positive; in absolute terms, they still represent a high amount due to the extremely high concentrations of total bacteria (Seedorf *et al.*, 1998). All the Gram negative bacteria found by Zucker *et al* (2000) in livestock houses are pathogenic. Endotoxins are lipopolysaccharide complexes originating from their outer membrane (Seedorf *et al.*, 1998), e.g. of *Escherichia coli, Salmonella, Shigella, Pseudomonas, Neisseria, Haemophilus*. Concentrations of endotoxins around livestock production areas are also high, found in the range between 0.66 and 23.22 endotoxin units (EU) m^{-3} (Schulze *et al.*, 2006). These concentrations can be easily exceeded inside livestock houses, reaching maximum values of 761 EU m^{-3} for dairy cows (Zucker and Muller, 1998), and 8120 EU m^{-3} for free-range hens (Spaan *et al.*, 2006). Lung infections by airborne fungi and airway-related inflammatory responses due to endotoxin exposures, are common animal respiratory diseases which are also related to impairment of the health and welfare of farmers and their neighbors (Auvermann *et al.*, 2006; Bakutis *et al.*, 2004; Radon *et al.*, 2002, Thorne *et al.*, 2009).

Pathogenic bio-aerosols can cause direct harm to the animals within livestock houses. Relatively long distance transmission of these pathogenic airborne bio-aerosols to vicinity farms can happen as well. For example, foot-and-mouth disease virus could be transmitted in the air and may infect animals in farms, some kilometers away from the source (Donaldson *et al.*, 1970; Hugh-Jones and Wright, 1970). When pathogens are zoonotic and airborne transmittable, health of farmers and people living nearby the livestock house may be challenged (Radon *et al.*, 2007; Wathes *et al.*, 2004). Table 2.1 lists some of the most relevant zoonoses, and micro-organisms responsible for them, which have been recovered from the air, thus they are potentially airborne transmittable.

Table 2.1. Potential airborne transmittable zoonoses.

Zoonoses	Micro-organism	Source
Campylobacteriosis	*Campylobacter spp.*	Berrang *et al.* (2004) Wilson (2004)
Influenza	Avian Influenza virus	Power (2005)
Newcastle disease	Newcastle disease virus	Hugh-Jones *et al.* (1973)
Colibacillosis	*Escherichia coli*	Sauter *et al.* (1981); Zucker *et al.* (2000)
Salmonellosis	*Salmonella spp.*	Gast *et al.* (2003)
Foot and mouth disease	Foot and mouth disease virus	Ryan *et al.* (2007)

Other bioactive components which can be found in livestock PM are antibiotics. Five different antibiotics were detected from PM in a pig house, at concentrations ranging from 0.2 to 12.5 mg kg^{-1} dust (Hamscher *et al.*, 2003). Tylosin, sulfametazine and tetracyclines were present and identified in the majority of the samples. These authors even identified traces of chloranphenicol, a prohibited substance since 1994.

2.3.2. Environmental hazards

There is a high potential of the livestock sector to contribute to PM emissions, and therefore, to environmental and climate related aspects. Environmental PM causes reduced visibility, vegetation stress and ecosystems alteration (Grantz *et al.*, 2003). Particulate matter can affect individual plants, plant populations, forest trees and terrestrial ecosystems. The majority of the documented toxic effects of particles on vegetation are

a reflection of their chemical components (acid/base, trace metals, and nutrients) (Grantz *et al.*, 2003).

Submicron particles scatter more light per unit mass and have a longer atmospheric lifetime than larger aerosols (IPCC, 2001). They have a direct radiative forcing because they scatter and absorb solar and infrared radiation in the atmosphere. Particles also alter warm, ice and mixed-phase cloud formation processes by increasing droplet number concentrations and ice particle concentrations (IPCC, 2001). A thorough review of this matter can be found in the United States' Environmental Protection Agency's report about environmental effects of airborne PM (EPA, 2004). The deposition of PM on surfaces may also cause damages by corrosion and deteriorate painted surfaces and other building materials (EPA, 2004). So far, the fate of PM emissions from livestock farms has neither been fully evaluated, nor directly related to such ecosystems alterations, although PM emissions from livestock houses can potentially contribute to such effects.

Near farms, particles which are released from livestock houses may also undergo rapid physical and compositional changes as they are emitted from the source, and transported away from it. These changes affect their size distribution and chemistry. This is particularly relevant to the formation rate of secondary inorganic particles in agricultural environments.

The formation of secondary inorganic particles in the presence of NH_3, as a result of chemical atmospheric reactions, is a major concern (Lammel *et al.*, 2004; Roumeliotis and Van Heyst, 2008). Ammonia can react with sulfuric (H_2SO_4), nitric (HNO_3), and hydrochloric acid (HCl) gases to form secondary inorganic particles such as ammonium sulfate (($NH_4)_2SO_4$), ammonium bisulfate (NH_4HSO_4), ammonium nitrate (NH_4NO_3), and ammonium chloride (NH_4^+Cl), which can be either solid or liquid (Robarge *et al.*, 2002). The contribution of secondary PM to total PM emissions from livestock houses, however, is still unclear. Although Roumeliotis and Van Heyst (2008) reported secondary inorganic particles could contribute to more than 50% of the total PM2.5 in a layer house, the origin of acidic species which can react with ammonia inside livestock houses remains unexplained.

In livestock environments, it can be assumed that there is always enough NH_3 inside and outside livestock houses. In the presence of excess NH_3 and high humidity, particulate NH_4^+ and NO_3^- could be formed. Other key factors such as particle mass, NH_3/ NH_4^+ balance, NH_4-NO_3 equilibrium, relative humidity, and temperature are also necessary to trigger secondary inorganic particle formation (Sharma *et al.*, 2007; Vogt *et al.*, 2005).

High NH_4^+ and NO_3^- salts, organic carbon, and calcium (Ca) in PM from livestock houses have been measured (Lammel *et al.*, 2004). Measured particle numbers 100 to 400 m downwind farms, were higher in the size ranges from 1 to 4 µm, and from 4 to 20 µm, than the background concentrations of particles. An increase in SO_4^{2-}, NO_3^-, NH_4^+, Ca, organic carbon, and elemental carbon in the PM10 fraction was found by Martin *et al.* (2008), also downwind from pig barns. The PM2.5 mass and number size concentrations also showed an increase which could be attributable to the farm (Martin *et al.*, 2008).

2.3.3. Legal framework to control PM in ambient air

There is lack of legislative background and framework for PM in Europe, although PM issues have found entrance into national and international regulations and strategies of air pollution and control. There have been attempts, however, to regulate emissions of PM, and moreover, emission inventories are in force.

European Directives "Council Directive 1999/30/EC relating to limit values for sulfur dioxide, nitrogen dioxide and oxides of nitrogen, PM and lead in ambient air", daughter directive of the "Air Quality Directive" (1996/62/EC), set limits to concentration of PM in ambient air, although these are seldom applied in livestock husbandry. Daily limit value for PM10 is 50 µg m^{-3}, not to be exceeded more than 35 days per year, and annual average limit is 40 µg m^{-3}. These limits became legally binding in 2005. The annual average limit would be reduced to 20 µg m^{-3} in 2010, although it seems it will be postponed to 2011, because of Europeans's lower level of ambition in this sense (Brunekreef and Maynard, 2008).

The new Air Quality Directive (2008/50/EC) has set an annual average limit for PM2.5 of 25 µg m^{-3} to be met by 2015. It has also set an annual

exposure concentration obligation of 20 µg m^{-3}, also to be met by 2015, based on an average exposure indicator measured on three consecutive years. Lastly, it has also set a national exposure reduction target for member states, where member states should reduce their PM2.5 concentrations by a certain percentage, also based on the average exposure indicator.

The Convention on Long-Range Transboundary Air Pollution provides an international policy framework to tackle atmospheric pollution problems, as well. There are several Protocols of the Convention that contain obligations for emission reductions that can also influence PM concentrations in ambient air, which are expected to lead to reduced ambient concentrations of PM in Europe (IIASA, 2006). These protocols require measures to reduce precursor emissions of secondary organic and inorganic particles, and prescribe technological standards that limit primary emissions of fine PM, in addition to other pollutants.

No specific legislation regarding maximum PM concentrations or emissions neither in agricultural environments nor in and from livestock houses is in force, despite the different policies and regulations. There are no maximum exposure limits set for livestock houses, although maximum levels have been determined experimentally in some cases. Considering the heterogeneous nature and complex mixtures of compounds found in PM, exposure thresholds are even more difficult to establish.

Donham *et al.* (1995) reviewed previously reported dose-response research in pig farm workers, resulting in exposure limit recommendations of 2.5 mg m^{-3} for inhalable PM, and 0.23 mg m^{-3} for respirable PM. For poultry workers, exposure concentrations associated with significant pulmonary function decrements were 2.4 mg m^{-3} for inhalable PM, and 0.16 mg m^{-3} for respirable PM (Donham *et al.*, 2000). Legally binding workplace exposure limits in the United Kingdom, are 10 mg m^{-3} for inhalable PM, and 4 mg m^{-3} for respirable PM, for an 8-hour average. For short term exposure (15 minutes), exposure limit is 20 mg m^{-3} for inhalable PM (HSE, 2007). The German Ordinance on Hazardous Substances (GefStoffV) also establishes short term (15 minutes) workplace exposure limits of 10 mg m^{-3} for inhalable PM, and 3 mg m^{-3} for respirable PM (BGIA, 2009). Recommended limits for animals are 3.4 mg m^{-3} for inhalable PM, and 1.7 mg m^{-3} for

respirable PM (CIGR, 1992), although these were changed to 3.7 mg m^{-3} for inhalable PM, and 0.23 mg m^{-3} for respirable PM, in a second review about recommended thresholds regarding pig health (CIGR, 1994).

Emission data to estimate local, national or international emissions of PM are still scarce, but PM inventories are now in focus, and annual PM emissions have to be calculated. Emission estimates are based on "emission factors". Fraction PM10 and PM2.5, at least should be measured, to comply with European legislation requirements. Furthermore, fractions PM4 and inhalable PM, can also be determined for other purposes.

Current estimates of PM emissions can be found in the national submissions under the Coordinated European Particulate Matter Emission Inventory project, initiated in 2000, under the Convention on Long-Range Transboundary Air Pollution. The European Pollution Emission Register also keeps records of PM emissions, for the different activities listed under the IPPC Directive (1996/61/EC), including poultry and pig farms. Estimates of PM emissions can also be obtained from the International Institute for Applied Systems Analysis's Regional Air Pollution Information and Simulation: RAINS model.

Emission factors currently used are, however, still far away from being capable of giving precise and reliable emission estimates. Emission factors must be determined by measurements or with the use of measurement aided models (Hinz, 2005). Efforts are still needed to improve the quality and precision of emission inventories in order to quantify more exactly emissions from livestock production systems, to establish adequate and accurate abatement strategies to reduce emissions.

2.4. Particulate matter in and from livestock production systems

2.4.1. Source apportionment of PM

In livestock production systems, so far, no exhaustive source apportionment studies have been carried out. Attempts to identify and quantify sources of PM in livestock houses, however, have been made. Main sources of PM have been identified in pig and poultry houses.

Research on sources of PM in livestock houses has focused on primary sources. In pigs, the bulk of PM comes from feed (Donham *et al.*, 1986; Heber *et al.*, 1988a; Honey and McQuitty, 1979; Takai *et al.*, 1998). Feed particles are more abundant in coarse fractions (Donham *et al.*, 1986). Curtis *et al.* (1975) found higher N content in aerial PM in a pig finishing house than that in settled dust or in the diet, indicating a possibly high contribution of feed to airborne PM, as well as of other sources, probably skin from animals, and other nitrogenous compounds in the air which could adhere to the suspended particles. Fecal PM is also important contributor to PM in pigs. Fecal PM is found in a greater extent in the respirable fraction, indicating a potential high risk to the alveoli in the lungs (Donham *et al.*, 1986). Pigs dander, mould, pollen and grains, insect parts, and mineral ash are less abundant (Donham *et al.*, 1986). Similar results were obtained by Heber *et al.* (1988a) in pig finishing house, with PM mainly coming from feed and to a lesser extent from feces. Size differentiated source analysis in pig PM, also showed about 5 to 10% of total PM were skin particles, being half in the size range from 7 to 9 μm (5%), compared with the size range from 11 to 16 μm (10%) (Honey and McQuitty, 1979). Animals themselves, however, were the main source of PM in another study (Nilsson, 1982). Aarnink *et al.* (1999) identified feed and skin particles being the most abundant sources in pigs.

In poultry, down feathers, mineral crystals from urine and litter, are the main sources in broiler houses (Aarnink *et al.*, 1999). In layer houses, skin, feathers, feces, urine, feed, and litter, are amongst the most important sources (Qi *et al.*, 1992). Other livestock production systems may reveal other relevant sources of PM different from the previous ones. For instance, bedding material can considerably contribute to PM, in particular systems where bedding is used, compared with non-bedding systems (Aarnink *et al.*, 2004). In growing pigs, inhalable concentrations of PM in systems where straw was used as bedding were doubled, compared with non-straw concrete floor systems, especially by the end of the fattening period, attributable to the use of straw. By the end of the fattening period, straw bedding became more "dusty" as it was more dirty, disintegrated, and could potentially generate more particles (Aarnink *et al.*, 2004). Type of litter and moisture content may also affect PM concentrations (Kaliste *et al.*, 2004).

In rabbits, fur, skin particles, feces, urine, feed, bedding, and disinfectants are major sources (Kaliste *et al.*, 2002). Feed, feces and disinfectants were identified by Cambra-López and Torres (2008) with scanning electron microscopy with X-ray single particle analysis (SEM-EDX) of rabbit house PM in different size fractions.

Source apportionment is a critical aspect in PM characterization. In ambient monitoring networks, source apportionment studies are very important. Source contributions of ambient PM concentrations have traditionally been estimated with dispersion models, using emission inventories for different sources as input data. Alternative techniques to determine source contributions are receptor models, or source apportionment studies, which use particle chemical characterization and chemical composition of PM collected at certain sites, instead. With source apportionment studies, composition of collected PM samples can then be related to the different sources.

There are two different approaches in source apportionment studies: general multivariate techniques and multiple linear regression models. When sources are well known and there are known chemical profiles, multiple linear regression models (e.g. chemical mass balance) can be applied, to determine the contribution of each source to sampled PM. If no chemical profiles are available, multivariate techniques can be used, which do not require known chemical source profiles, but require a high number of samples to be analyzed (Watson *et al.*, 2002).

Morphology and chemical composition of PM in livestock houses, however, with high organic content, makes the application of source apportionment as applied in ambient air difficult, where many inorganic elements can be distinguished and apportioned to a certain source (Almeida *et al.*, 2005; Almeida *et al.*, 2006). Morphology of the most abundant organic particles in livestock house PM sources (feed and feces) can be very similar. To discriminate feed particles from fecal particles or undigested feed particles is thus difficult, especially when only light microscopy is used. Results in some studies, therefore, tend to overestimate the contribution of particles from feed, because no distinction is made between feed and feces, as in the study conducted by Honey and McQuitty (1979). The use of stains might be

helpful in this case, like in Donham *et al.* (1986), where iodine was used to stain starch from feed particles, and nile blue sulfate to stain fecal particles.

Chemical composition in livestock PM shows high contents of C, O, N, P, S, Na, Ca, Cl, Mg, and K in most contributing sources (see Chemical composition of PM). Chemical composition of feed, fecal, and skin particles showed relatively similar concentrations of N, K, Cl, and Na in skin particles compared with feed and feces, although higher concentrations of P in fecal particles compared with feed or skin particles (Aarnink *et al.*, 1999). Chemical composition of particles using SEM-EDX can be moreover, a difficult task. The SEM-EDX spectrum for skin particles shows a high percentage (>5%) of C and O, minor (1-5%) of Si, and very little (<1%) of Cl, Na, P, S and Ca; whereas feed particles present a spectrum with high percentage (>5%) of C and O, minor (1-5%) of Si, Cl, P, Ca, and very little (<1%) of Mg, Al, Na (Cambra-López and Torres, 2008; McCrone, 2007). Some elemental spectrum obtained with SEM-EDX of PM are shown in Figure 2.4, for skin and feed particles, showing similarities in main elements.
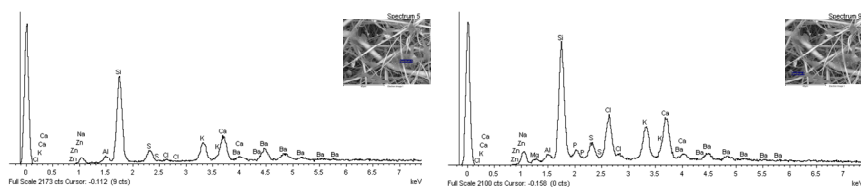
Figure 2.4. Scanning electron microscopy with X-ray single particle analysis for skin particle (photograph on the left) and feed particle (photograph on the right). From Cambra-López and Torres (2008).

2.4.2. Particulate matter levels: Concentration and emissions

The formation of PM, its concentrations and emissions from livestock houses depends on many physical and biological factors. As research has focused mainly on poultry and pigs, most published information refers to these species.

Available data on inhalable and respirable PM concentrations are shown in Table 2.2 (for broiler houses), Table 2.3 (for layer houses), and Table 2.4 (for pig houses). Note that inhalable PM is considered equivalent to TSP, and respirable PM in these cases is used equivalent to PM4 (according to EN 481:1993 and ISO 7708:1995), or PM5 (50% cut-off diameter of 5 μm, according to fine dust definition of the Convention of Johannisburg).

Concentrations of PM are higher in poultry than in pigs, and higher in broilers (litter systems) than in hens (cages). For other poultry such as turkey production, concentrations of TSP are comparable with inhalable PM concentrations for broilers shown in Table 2.2. Concentrations of TSP in turkey houses ranged from 1.3 to 7.5 mg m^{-3} (Hinz *et al.*, 2007a). For cattle, concentration measurements suggest PM in these houses is low. Total suspended PM concentrations in a dairy cattle house have been found below 1 mg m^{-3} (Hinz *et al.*, 2007b).

Emissions are more difficult to calculate and therefore, there is also severe lack of data. Only limited number of studies has reported on emission factors for PM from livestock houses. These data are necessary, however, for decision-making and evaluation of emission control strategies, as well as for monitoring of atmospheric pollution, to assess the impact of livestock operations on the surrounding environment. Emission rates from livestock buildings are, furthermore, difficult to compare due to the use of non-standardized and homogeneous units amongst species, usually expressed per animal or live weight.

The reliability of some of these emission data is also questionable, because emission factors must be derived from research. The quality of these is improved when the methodology which is used is sound, the number of sources tested and the sources themselves are representative, and the results are presented in such a way that it permits validation (NRC, 2002). To date, the work performed by Takai *et al.* (1998) still represents one of the most comprehensive and exhaustive studies conducted in livestock buildings in Europe, in PM emission rates.

Table 2.2. Review of measured inhalable and respirable PM concentrations in broiler houses with litter in chronological order of publication.

	Concentration (mg·m⁻³)		Country	Source
	Mean	Range		
Inhalable PM	10.1	-	England	Wathes *et al.* (1997)
	-	9.2 - 11.1	Scotland	Al Homidan *et al.*(1998)
	-	1 - 14	Germany	Hinz and Linke (1998b)
	7.15	3.83 - 10.36	England, The Netherlands, Denmark, and Germany	Takai *et al.* (1998)
	3.21	-	The Netherlands	Aarnink *et al.* (1999)
	-	8.2 - 9	The Netherlands	Ellen *et al.* (1999)
	-	0.73 - 11.39	U.S.	Redwine *et al.* (2002)
	-	1.77 - 4.41	Scotland	Al Homidan (2004)
	4.32	2.27 - 8.58	Australia	Banhazi *et al.* (2008)
	-	2.0 - 4.9	Croatia	Vucemilo *et al.* (2008)
Respirable PM	5.43 (28th day) 9.71 (49th day)		U.S.	Willis *et al.* (1987)
	0.10	-	England	Wathes *et al.* (1997)
	0.81	0.42 - 1.14	England, The Netherlands, Denmark, and Germany	Takai *et al.* (1998)
	-	1.4 - 1.9	The Netherlands	Ellen *et al.* (1999)
	0.84	0.30 - 1.8	Australia	Banhazi *et al.* (2008)

Table 2.3. Review of measured inhalable and respirable PM concentrations in layer houses with or without litter, in chronological order of publication.

	Concentration (mg·m⁻³)		Country	Housing system	Source
	Mean	Range			
Inhalable PM	1.7	-	England	Cages	Wathes *et al.* (1997)
	2.8	-		Litter (perchery)	
	1.22	0.75 - 1.64	England, The Netherlands, Denmark, and Germany	Cages	Takai *et al.* (1998)
	5.28	2.19 - 8.79		Litter (perchery)	
	-	1.2 - 5.5	Sweden	Litter	Gustafsson and von Wachenfelt (2006)
Respirable PM	0.10	-	England	Cages	Wathes *et al.* (1997)
	0.17	-		Litter (perchery)	
	0.14	0.03 - 0.23	England, The Netherlands, Denmark, and Germany	Cages	Takai *et al.* (1998)
	0.84	0.35 - 1.26		Litter (perchery)	

Table 2.4. Review of measured inhalable and respirable PM concentrations in pig houses, in chronological order of publication.

	Concentration (mg m⁻³)		Country	Animal type	Source
	Mean	Range			
Inhalable PM	6.25	3.2 - 15.3	U.S.	Pigs (diverse)	Donham *et al.* (1986)
	7.8	3.1 - 14.5	U.S.	Finishers	Heber *et al.* (1988b)
	0.72	0.12 - 2.14	U.S.	Fatteners	Maghirang *et al.* (1997)
	-	1 - 5	Germany	Fatteners	Hinz and Linke (1998b)
	2.19	1.87 - 2.76	England, The Netherlands, Denmark, and Germany	Sows, weaners and fatteners	Takai *et al.* (1998)
	0.43	-	Netherlands	Weaners	Aarnink *et al.* (1999)
	-	0.79 - 1.91	Sweden	Fatteners-finishers	Gustafsson (1999)
	-	0.42 - 6.86	U.S.	Finishers	Schmidt *et al.* (2002)
	-	2.08 - 5.67	The Netherlands	Fatteners-finishers	Aarnink *et al.*(2004)
	-	0.05 - 5.6	U.S.	Finishers	Bottcher *et al.* (2004)
	-	0.4 - 3.7	Germany	Fatteners	Haeussermann *et al.* (2006)
Respirable PM	0.23	0.18 - 0.26	England, The Netherlands, Denmark, and Germany	Sows, weaners and fatteners	Takai *et al.* (1998)
	-	0.09 - 0.30	Sweden	Fatteners-finishers	Gustafsson (1999)
		0.04 - 0.44	U.S.	Finishers	Schmidt *et al.* (2002)
	-	0.16 - 0.71	The Netherlands	Fatteners-finishers	Aarnink *et al.* (2004)
	-	0.00 - 0.85	Germany	Fatteners	Haeussermann *et al.* (2006)

2.4.3. Factors affecting PM generation and levels

Particulate matter formation and concentration depends on factors related to the kind of housing and feeding, animal type, and environmental factors. Concentrations of PM in livestock houses mainly depend on animal type, as well as on housing system, season and sampling period within a day (Ellen *et al.*, 2000). Animal activity, animal density and moisture conditions are also important. A relative humidity of 70% or higher may contribute to low PM concentrations due to a high equilibrium moisture content. Above this moisture content, Takai *et al.* (1998) indicated that particles may contain bound and condensed water, which may cause particles to aggregate together. Investigations in a broiler house have shown total PM concentrations are significantly influenced by temperature and relative humidity (Vucemilo *et al.*, 2008).

Models have been used to predict relationships between PM and other related factors. Haeussermann *et al.* (2008) recently developed a dynamic model to predict PM10 emissions in pig houses. Ventilation rate, animal activity, feeding operations, indoor humidity, animal weight and finishing day were amongst all the main influencing factors on PM10 concentrations and emissions. Housing system considerably influenced PM10 concentrations and emissions in this study. By multi-factorial linear analysis Banhazi *et al.* (2008) identified statistically significant factors influencing inhalable and respirable PM concentrations in broiler houses. This analysis showed on one hand that the type of ventilation rate, type of bedding, temperature, and building age significantly influence inhalable PM concentrations; and on the other hand, that respirable concentrations were more influenced by cleaning or not cleaning between batches of birds, number of birds per building airspace, ventilation levels and humidity.

In practice, with experimental studies, similar factors influencing PM formation, concentration, and emission in and from livestock houses as those identified using dynamic model approaches and multi-factorial linear analysis, have been identified. However, correlation between some of these factors, (e.g. temperature and ventilation) difficulties attribution of PM formation, concentration, and emission to a single factor. Inhalable and respirable PM, moreover, behave differently, presumably because factors influencing inhalable and respirable PM can differ not only in formation, but also in concentration and emission processes. It is still not clear how temperature and relative humidity for instance, influence PM concentrations. Ventilation rate, also related to temperature and relative humidity, is an important factor, because it determines to a great extent particle concentrations and emissions, and especially its distribution in the airspace of livestock houses (Puma *et al.*, 1999b; Puma *et al.*, 1999a). Amongst factors influencing PM, however, animal type and housing system are critical, because they are intrinsically related to other influential factors such as feeding operation, ventilation rate, and animal activity. Other critical factors such as animal number and age, also weight, are furthermore related to ventilation rate, temperature and relative humidity.

Amongst all poultry houses, poultry raised on litter show higher PM concentrations than poultry in caged systems (Takai *et al.*, 1998). In pig

houses, higher PM concentrations are found in systems with bedding than in those with concrete floors (Aarnink *et al.*, 2004). Type of litter and moisture content may also affect PM concentrations (Kaliste *et al.*, 2004).

Bird age has a positive relationship with PM concentrations and emissions rates (Redwine *et al.*, 2002). Hinz and Linke (1998b) determined PM concentrations increased linearly with broilers live weight. Yoder and Van Wicklen (1988) found a logarithmical relationship between respirable PM concentration and total bird weight. Bird age affects PM emissions from broilers probably because of changes in litter moisture and composition (Wathes *et al.*, 1997). Increased amount of dried manure with bird age, as well as increased bird activity, and ventilation rates may also contribute to increased PM emissions. On the contrary, in pigs, average inhalable PM concentrations decreased with live weight, probably due to reduced animal activity as pigs grew (Hinz and Linke, 1998b). Increased ventilation rate with pig live weight may have also caused reduced PM concentrations as pigs grew.

Climate or season, which is intrinsically related to ventilation rates, also affects PM concentrations inside farms. Because of higher ventilation rates in summer compared with winter, low PM concentrations and high emission rates can be expected in the summer, whereas high PM concentrations and low emission rates can be expected in winter (Hinz and Linke, 1998b; Redwine *et al.*, 2002). In a layer house, total and respirable PM formation rates were higher during hot weather ventilation rates, than during cold weather ventilation rates for total and respirable PM, because of an increased turbulence and suspension of PM (Qi *et al.*, 1992). In the study by Takai *et al.* (1998), the effect of season on inhalable and respirable PM concentrations was significant in all cases, for both pig and poultry farms. Concentrations were higher in winter for both fractions. In a broiler house PM concentrations were lower when the inside temperature exceeded the outside temperature by less than 10ºC (summer), than when the difference was more than 10ºC (winter) (Hinz and Linke, 1998b). Smaller differences between outside and inside temperature are normally associated with higher ventilation rates, thus showing again a high influence of ventilation rates on PM concentrations.

The application of lighting programs results in changes in PM concentrations (Ellen *et al.*, 2000). Particle formation rates in a poultry layer house were significantly higher during light periods than during dark periods (Qi *et al.*, 1992). Yoder and Van Wicklen (1988) also reported higher mean respirable PM concentrations during light periods than during dark periods in broilers. These authors related this difference to the observed increased bird activity during light periods. Sampling period within a day and the use of lighting programs is, therefore, critical and affects PM formation and concentrations in relation to animal activity. Farm animals are generally more active during daytime because feeding, and activities of farmers, are mainly restricted to the daytime. Animal movement causes turbulence around them and disperses settled PM from building surfaces, causing an increase in PM concentration (Takai *et al.*, 1998). This effect may differ amongst particle sizes, because coarse particles tend to settle quicker than smaller and finer particles, which tend to remain airborne for longer times.

In hens, because of greater laying hen activity during the day, PM concentrations seem to be more related to day/night changes than in broilers (Takai *et al.*, 1998; Wathes *et al.*, 1997). In broilers, however, most studies were conducted under 24-hour lighting programs (Hinz and Linke, 1998a; Redwine *et al.*, 2002; Wathes *et al.*, 1997), which have almost disappeared nowadays. In broilers with intermittent lighting programs, there appears to be a strong relationship between the lighting program, animal activity and the concentration of PM, as determined by Calvet (2008) when assessing PM10 concentrations in relation to measured bird activity through an activity bird index (Figure 2.5). In pigs, inhalable PM concentrations increased at feeding times, and thus concentrations were higher during daytime than at night (Hinz and Linke, 1998b). With a dry feeder, feeding twice a day has shown lower PM concentrations than free access to feed in pigs (Bundy and Hazen, 1975). These results also suggest an influence of animal activity related to PM emissions. Farm activities, sunlight entering the building, sunrise or sunset, as well as any factor which may cause animal excitement and animals to be more active, seem to be a main cause for increased amounts of PM being formed, and also becoming airborne.
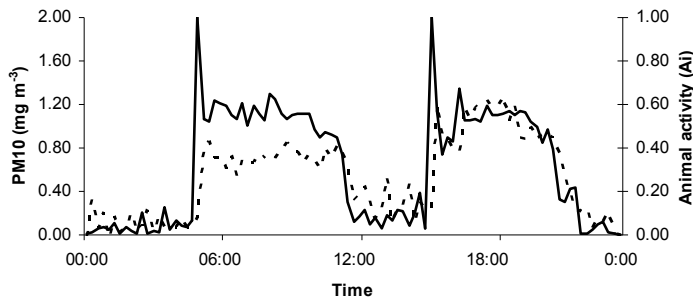
Figure 2.5. Relationship between PM10 concentration (dotted line) and animal activity (Ai) (continuous line) in broilers, expressed as weekly averages, during the third week of the rearing period. Note dark periods from 21:00 to 05:00, and from 11:30 to 15:30. From Calvet (2008).

A difference can be made between processes leading to the formation of PM and processes leading to PM becoming airborne (Aarnink and Ellen, 2007). Processes leading to formation of PM are influenced by processes inside and outside the livestock house. Processes leading to PM becoming airborne are mainly influenced by animal and human activities. To be able to develop PM reducing measures for livestock houses, sources of PM and the processes leading to PM formation and emission must be understood in any specific situation (Aarnink and Ellen, 2007). These stages in PM formation and dynamics are illustrated in Figure 2.6, which reviews the main processes and factors involved. Once these processes and factors are known and understood, this schematic picture can help to decide where to focus and where to act to reduce PM formation and emissions.

2.4.4. Particle size and size distribution

Particle-size distributions in livestock houses can be very variable. It is important when assessing PM reduction techniques, to identify which particle size ranges remain, and which are more effectively removed (Dawson, 1990). These PSDs can furthermore change in time. Mean particle size increased with bird age in broilers (Yoder and Van Wicklen, 1988).
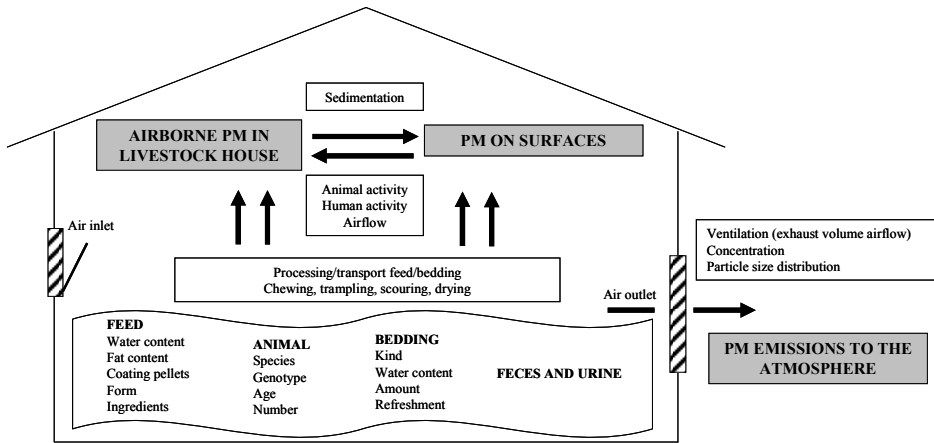
Figure 2.6. Schematic overview of PM processes and factors leading to PM formation and emission.
Adapted from Aarnink and Ellen (2007).

Particulate matter emissions in livestock houses are both fine and coarse. The percentage of coarse PM of TSP exceeds 85% for most livestock species (Romann and Hinz, 2007). Table 2.5 reviews different size fractions reported in the literature for livestock farming showing how fraction larger than PM10 is generally higher than the rest. The PM10 fraction is partly composed of PM2.5. Roumeliotis and Van Heyst (2007) reported a contribution of PM2.5 to PM10 of about 75%. Submicron particles, those in the PM1 fraction, have seldom been measured. Preliminary results show their concentrations in livestock houses are not negligible (Roumeliotis and Van Heyst, 2007).

Table 2.5. Size fractions reported in the literature for livestock species, as percent of total suspended particles. Adapted from IIASA (2006).

Sector	PM2.5	PM5	PM10	> PM10
Pigs	8 - 12%	4 - 14%	40 - 45%	55%
Broilers	9%	-	58%	42%
Laying hens	3%	-	33%	67%
Cattle	-	17%	-	-

Most PSDs of livestock PM are reported as bi-modal (Schneider *et al.*, 2001; Schneider *et al.*, 2006). This bi-modal distribution can partially be explained because emissions of primary particles fall into the coarse mode, whereas emissions of secondary particles fall into the accumulation mode. Primary organic particles, however, have also been identified in the accumulation

mode, together with secondary sulphate background PM in livestock houses (Schneider *et al.*, 2001).

Both mass and number distributions have been evaluated and parameterized in terms of d_{50} and σ_g. Schneider *et al.* (2006) found in one mode, d_{50} were close to 0.30 μm, with σ_g of 1.45 μm, similar for all livestock species measured. In the next mode, d_{50} was close to 1.60 μm, with σ_g of 1.80 μm, higher than in the previous mode. Highest d_{50} values were found in pigs and poultry farms, for mass and number concentrations (Schneider *et al.*, 2006).

Numerical size distributions show very high numbers in the lowest size ranges. Depending on which measurement device is used, and the low detection size range, results may differ. When ultra-fine particles are measured, very high numbers are found. In the respirable fraction, however, small particles with diameters between 1 and 2 μm are most abundant in number, in broilers (Yoder and Van Wicklen, 1988). In laying hens houses, 99% of total number of particles was smaller than 10 μm in diameter, and 97% of total number of particles were smaller than 5 μm in diameter (Maghirang *et al.*, 1991). More than 40% of the total number of particles were in the range from 0.3 to 0.5 μm in diameter (Maghirang *et al.*, 1991).

Mass size fractions behave differently. Mass median diameters between 11 and 17 μm are usual found in pig and poultry houses (ICC and SRI, 2000). In broilers, these values can even be higher, between 24 and 27 μm (Hinz and Linke, 1998b). Broiler PM with litter exhibits higher d_{50} than PM from other housing systems without litter. The relative mass of small particles was found to be higher in rearing pigs compared with broilers (Aarnink *et al.*, 1999). Particles smaller than 5.8 μm were 29% of total mass of PM in pigs, but only about 18% in broilers (Aarnink *et al.*, 1999). In turkeys, 50% of total mass was attributable to particles larger than 53 μm (Hinz and Linke, 2006).

2.4.5.   Chemical composition of PM

Particulate matter from livestock houses consists up to 90% organic matter (Aarnink *et al.*, 1999; Seedorf and Hartung, 2001). It is mainly composed of primary particulates of biological origin, directly emitted from livestock husbandry, containing micro-organisms (fungi, bacteria, viruses, toxins and

allergens), and other substances such as feed, skin particles and fecal particles (Donham *et al.*, 1986).

Composition of PM mainly varies with animal species, and is intrinsically related to housing systems, and especially to the presence and composition of litter (Aarnink *et al.*, 1999). For instance, PM from poultry and pig houses is rich in N content and generally shows higher dry matter (DM) contents than PM from cattle barns, which is usually more humid and shows a higher content of minerals or ashes (Hartung and Saleh, 2007). Table 2.6 shows typical chemical composition of airborne PM in a pig house and in a broiler house, showing high DM and N contents, as well as relatively high P and potassium (K) concentrations compared with other analyzed elements. Nitrogen concentrations are higher in broilers (169 g/kg PM) than in pigs (67 g/kg PM).

Table 2.6. Chemical composition of airborne inhalable PM in a pig and broiler house by bulk analysis. From Aarnink *et al.* (1999).

	Dry matter (DM)	Ash	N	P	K	Cl	Na
			(g/kg)				
Rearing pigs	920	149.5	67.0	14.7	27.8	7.8	8.2
Broilers	911	97.4	169.0	6.4	40.3	4.2	3.2

Bulk analysis and single particle analytical techniques have been used to evaluate PM chemical composition in livestock houses. Most inorganic elements in PM can be analyzed with several techniques which differ in detection limits, sample preparation, and costs. Some of these techniques are non-destructive and conserve PM samples for further analysis (e.g. energy dispersive X-ray fluorescence and proton induced X-ray emission), and can detect more than 40 elements with relatively low detection limits. Elemental carbon, organic carbon or carbonate carbon in PM are usually determined by thermal-optical techniques. Some techniques can determine total carbon (sum of elemental, organic and carbonate carbon), from which the chemically different carbon forms can be estimated by subtracting the corresponding amounts. Other techniques measure dissolved ionic elements, thus require samples and PM to be dissolved for analysis (e.g. atomic absorption spectrophotometry, inductively coupled plasma with atomic emission spectroscopy, and inductively coupled plasma with mass

spectroscopy), and can also detect many elements that can be dissolved (Martin *et al.*, 2008).

Organic particles are more often analyzed by means of light microscopy and scanning electron microscopy. Electron microscopy is a very powerful tool for PM analysis because it gives higher magnification than light microscopy with three dimensional shape-resolved images, which enables better identification and physical characterization of single particles. Scanning electron microscopy with X-ray single particle analysis adds to particle physical characterization, chemical characterization, and the possibility of elemental analysis. This technique has been extensively used for PM analysis, but requires a high number of particles to be analyzed to obtain statistically sound results, and thus is time and labor consuming (Mamane *et al.*, 2001).

Scanning electron microscopy with X-ray analysis was carried out in PM collected from different housing systems for growing pigs. Area measurements of certain spots in PM samples, instead of single particle analysis, were done. Samples showed Na, magnesium (Mg), Al, P, S, Cl, K, and calcium (Ca) were the most abundant elements (Aarnink *et al.*, 2004). Schneider *et al.* (2002) also found high contents of P, N, K, and Ca in particles from a piggery in different size ranges. This is a novelty, because PM chemical composition in livestock houses has seldom been analyzed discriminating size. Small particles (about 0.65 µm in diameter) were rich in S, O, and C, corresponding to secondary SO_4^{2-} particles, and the rest were particles of biological origin (Schneider *et al.*, 2002). Fecal particles showed a typical carbonaceous composition and strong presence of P, with high organophosphate and pyrophosphate signals (Schneider *et al.*, 2002).

2.5.   Abatement strategies in livestock production systems

Because PM emissions from livestock houses are strongly related to kind of housing and feeding, animal type, and environmental factors, abatement strategies must preferably be site specific. However, also less site specific techniques such as air scrubbers can very efficiently reduce PM emissions. Most of the reduction options follow directly from the processes and factors that have been discussed in Figure 2.6. Concentrations and emissions of PM emissions can therefore be reduced in any of the steps

described in Figure 2.6. Research has given ground to the development of a wide range of PM reduction strategies, which have proven to be efficient in livestock systems: "low-dust" feed (Dawson, 1990; Nannen *et al.*, 2005), de-dusted bedding material, use of feed additives (Guarino *et al.*, 2007; Takai and Pedersen, 2000; Zhang *et al.*, 1995), water or oil sprinkling (Takai and Pedersen, 2000; Zhang *et al.*, 1995), changes in ventilation rate and air distribution (Aarnink and Wagemans, 1997; Gustafsson and von Wachenfelt, 2006), vacuum cleaning (Nilsson, 1982), end-of-pipe techniques (filtration and wet scrubbing) (Bottcher *et al.*, 2000; Dawson, 1990; Kosch *et al.*, 2005; Melse and Ogink, 2005; Willis *et al.*, 1987; Zhang *et al.*, 2005), and electrostatic precipitation and ionization (Chiumenti and Guercini, 1990; Dolejs *et al.*, 2006; Mitchell *et al.*, 2000; Mitchell *et al.*, 2004; Ritz *et al.*, 2006; Rosentrater, 2003; Tanaka and Zhang, 1996).

These strategies can be classified into three main groups: source-control techniques which aim at reducing PM emission from the source, dilution and effective air room distribution, and PM removal or air cleaning techniques (e.g. scrubbers, ionizers or electrostatic precipitators) (Amuhanna, 2007). Special attention is given to the abatement strategies inside the houses, as regards to the end-of-pipe techniques, arguing that end-of-pipe techniques do not improve air quality inside livestock houses. End-of-pipe techniques, however, have been proven to be very efficient in reducing PM emissions to the environment (Ogink *et al.*, 2008; Ogink and Aarnink, 2007; Zhao *et al.*, 2008). Other techniques such as air ionization, oil spraying or simply changes in farm management can be more effective for this purpose (improving air quality inside and outside) (Aarnink *et al.*, 2008; Dolejs *et al.*, 2006). Efficiency of these techniques has been reviewed and discussed (Patterson and Adrizal, 2005; Pedersen *et al.*, 2000; Takai and Pedersen, 2000). Their application, however, to particular livestock houses needs to be further investigated, as well as their effect on emission reduction. Their effects on the different PM sources, in addition to particle sizes and PSDs, are still not clear.

There is still a long way ahead in order to establish the most efficient, technical, and economically viable abatement strategies, appropriate for particular livestock system, animal type, and geographical regions. Further research should address how to improve air quality inside and outside

livestock houses, combining different measures to tackle not only PM, but other pollutants such as ammonia and odor, as well. In this sense, the use of Computational Fluid Dynamics tools (CFD) can also be an appropriate method to optimize air flow patterns in livestock houses which can result in better air quality inside and reduced emission from the livestock house (Rong *et al.*, 2008).

2.6.   Research needs and future priorities

Livestock production systems are still one of the most poorly characterized sources in terms of pollutants and emissions. Two main areas of deficiency are evident: PM characterization and source apportionment; and PM levels and factors influencing these.

Although there are studies which have addressed PM characterization and source apportionment in literature (see Source apportionment of PM), most of them provide limited data from specific production systems related to single livestock categories. The results from these studies are derived from comparison of collected particles to known reference materials, but are generally based on light microscopy and visual counting, resulting in semi-quantitative estimations of source contributions, which can only account for particles bigger than 5 μm. These studies are therefore valuable for identifying the most likely sources present in different livestock production systems, but are far from providing comparable source contributions between and within livestock categories for different sized-particles. To this end, specific methodologies which include statistical methods to calculate source contributions and measuring protocols to characterize the morphology and composition of PM in different size fractions need to be developed. Peculiarities of PM in livestock houses: high organic content, adsorbed compounds, and high micro-organism content determine these methodologies. Because of high organic content of PM, and a high content of C, H, O, N, P, S, Na, Ca, Cl, Mg and K in most particles from feed, skin, and feces, it is important to distinguish the proportions of each inorganic element present in each source, which can aid in source identification and apportionment. A detailed profile of chemical composition of different PM sources is thus needed, to identify major PM contributors to tackle PM pollution at source. Other elements present in PM such as elemental carbon,

and ions can also be useful to identify sources different from mechanical fracture of feed, skin or fecal particles, such as combustion particles or secondary inorganic particles. Furthermore, comprehensive field studies which examine these characteristics are necessary.

Standardized measurement devices and measurement protocols to quantify PM levels in livestock houses also need further research. The validation and adaptation of PM sampling equipment to measure PM in livestock houses is necessary (Zhao *et al.*, 2009). Concentration and emission measurements should include monitoring of particle size and PSDs, as well. Identification of PSDs in different housing systems and animal types is needed. Size fractional analysis, furthermore, is also relevant to source apportionment and size segregated samples should be included in source apportionment studies.

To reduce potential health effects of PM, the absorbed gaseous and odorous compounds within PM is also an interesting area of research, especially with respect to NH_3 and odorous compounds. Reducing micro-organisms and bioactive compounds (endotoxins, antibiotics, allergens, dust mites and beta-glucans) in PM is also relevant to tackle health effects. The likely exposure of the population to PM, and the possible health effects of PM from livestock houses depend on the physical, chemical and microbiological characteristics of the emissions (Harish *et al.*, 2005; Wathes *et al.*, 2004). The direct cause-effect evidence of PM and its compounds on human and animal health are not yet clear.

To achieve accurate PM emission factors, relevant to different management practices, environmental parameters, and animal types, much more data is needed on emission rates. These data are needed to determine better mean emission rates and reliable emission factors. Particularly, research should focus on management variation factors related to feed, and litter composition and type, being these factors precisely related to geographical regions. Other factors related to animal activity, and lightning schemes, are equally relevant and must be investigated.

In this sense, investigations on morphological and chemical properties of the emitted PM, especially near the source, are lacking. To date, most studies have dealt with morphological and chemical properties of PM inside

livestock houses, but these particle properties, size distributions, and fate of PM in the environment, are still missing and neither sufficiently studied nor understood. Morphological and chemical composition of PM emitted from livestock houses would be especially valuable when addressing the problem of secondary inorganic particle formation, in terms of its influence on local and regional air quality. These findings will help understand the climate relevance and influence of livestock PM to atmospheric chemistry by means of systematically characterizing PM emitted from livestock houses. Such type of data can be also useful for modeling exercises, and to refine and develop more effective modeling tools, as well.

Some investigations show exceedance of the daily limit value for PM10 of 50 µg m^{-3} seems unlikely near livestock houses. Monitoring intensive poultry rearing operations did not show exceedance of this PM10 limit (Bull, 2008). Nevertheless, twenty-four-hour measurements of PM2.5 concentrations outside a broiler house ranged from 5.4 to 55.1 µg m^{-3} (Visser *et al.*, 2006). However, these studies suggest it is difficult to study PM emissions from livestock separately from other activities or sources which influence background PM levels in the area. Questions arise: should we shift the focus to fine PM, and what is the magnitude of secondary particle emissions related to farming activities? What fraction of total PM2.5 emissions corresponds to secondary particles? Such aspects are critical, and therefore, they must be looked at in depth, to relate and evaluate environmental hazards of PM from livestock houses.

In any case, the potential of livestock houses to PM2.5 and PM1 emissions is presumably high, but still unknown. PM1 fraction measurements should also be considered in monitoring programs and possibly in future regulations because the magnitude of this fraction might not be negligible (Roumeliotis and Van Heyst, 2007). Thus differential particle sampling is encouraged, even going down to the ultra-fine fraction.

In summary, research should address: particle composition and sources, particle size, PM levels and the factors influencing these, to provide the necessary information to stakeholders and local authorities involved, before decisions are made and effective abatement strategies are identified and implemented.

2.7.   Conclusions - Recommendations

1. Livestock production systems can emit considerable amounts of PM, which have to be controlled and reduced to protect the environment, and the health and welfare of humans and animals, and to comply with current European legislation on air quality.

2. Environmental and health effects of PM are strongly related to morphology, composition and levels of PM. Information on PM morphology and composition especially, is needed to identify and quantify main sources of PM, to evaluate its effects, and to propose adequate abatement measures.

3. Livestock husbandry is one of the more poorly characterized sources, and data on PM morphology and composition is limited, as well as on PM levels and factors influencing these levels.

4. Particulate matter from livestock houses is mainly coarse, primary in origin, and consists up to 90% organic matter. The relative contribution, however, of primary and secondary sources to PM is unclear. A detailed characterization and source apportionment of PM from different livestock houses is missing. Specific methodologies which include statistical methods to calculate source contributions and measuring protocols to characterize the morphology and composition of PM in different size fractions need to be developed. Comprehensive field studies need to be performed, as well.

5. The extent to which PM from livestock houses can adsorb and contain irritating gases such as NH_3, odorous compounds, and pathogenic and non-pathogenic micro-organisms is still uncertain. Attached to PM, these compounds, plus other bioactive components can enhance the biological effect of PM, and aggravate the potential health hazard.

6. Levels of PM (concentrations and emission) have been mainly measured in poultry and pig houses. Processes and factors involved in PM levels have been extensively investigated. Levels of PM are highest in broiler houses compared with other animal species,

probably attributable to the use of litter. Factors affecting PM levels in livestock houses are related to kind of housing and feeding, animal type, and environmental factors. More efforts, however, are needed to standardize measurement devices and protocols, to evaluate PM concentrations and emissions in relation to other animal types different from poultry and pigs, in different geographical regions.

7. Abatement strategies to reduce PM in and from livestock production systems are available, but to optimize the efficiency, and technical and economical viability of some of the most promising techniques such as air ionization, oil spraying, and changes in management, further research and adaptation to specific farm situations is required.

8. Control of PM in and from livestock production systems is not only a major challenge for modern livestock production, but also for stakeholders (farmers and local authorities), and for the scientific community, which can help fulfill the gap on knowledge with respect to: particle size and composition, PM sources, levels, and the factors influencing these.

2.8.   Acknowledgements

2.9.   References

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Chapter 3

Source analysis of fine and coarse particulate matter from livestock houses

Source analysis of fine and coarse particulate matter from livestock houses

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Abstract. The analyses of the different sources which can contribute to particulate matter (PM) emissions from livestock houses are essential to develop adequate reduction techniques. The aim of this study was to morphologically and chemically characterize several sources of PM from livestock houses. We collected known sources of PM from different housing systems for poultry and pigs, which were later aerosolized in a customized laboratory dust generator to collect fine and coarse PM samples. These samples were morphologically and chemically characterized using scanning electron microscopy with X-ray microanalysis to develop comprehensive morphological and chemical source profiles. Moreover, source particle-size distribution was determined. Results showed distinct and unique particle morphologies in collected sources from different housing systems for poultry and pigs. Although presence of N, Na, Mg, Al, Si, P, S, Cl, K, and Ca were identified in all sources, their relative elemental concentrations varied amongst sources and could be used to discriminate amongst them. Particle size and size distribution also varied amongst sources (size ranged from 2.1 to 18.1 μm projected area diameter), and mainly depended on its mineral or organic origin. Consequently, a comprehensive particle characterization and complete source analysis was achieved. The outputs of this work can be useful information for source identification and quantification in PM from livestock houses, improving the understanding of how PM is generated in such environments, which is essential for developing strategies for its reduction.

3.1.   Introduction

High concentrations of particulate matter (PM) can threaten the environment as well as the health and welfare of humans and animals. A close relation between PM air pollution, respiratory and cardiovascular disease, and mortality has been identified in the long term (Dockery *et al.*, 1993; Pope *et al.*, 2002), as well as in the short term (Ballester *et al.*, 2002; Hoek *et al.*, 2000). Particulate matter air pollution can also cause reduced visibility, vegetation stress, and ecosystems alteration (Grantz *et al.*, 2003). Furthermore, small PM can have a direct radiative effect because they scatter and absorb solar and infrared radiation in the atmosphere (IPCC, 2001). Data on particle morphology and chemical composition are essential to understand particle origin and fate, thus health and environmental hazards (Mamane *et al.*, 2001). Because these data cannot be inferred from mass measurements alone (Shi *et al.*, 2003), characterization of particle properties offers the potential to specifically identify and quantify sources of PM (Casuccio *et al.*, 2004).

Livestock houses are important contributors to ambient fine (PM2.5) and coarse (PM10-2.5) PM emissions (Takai *et al.*, 1998). Inside livestock houses, numerous studies have reported higher prevalence of respiratory diseases in livestock farmers compared with other occupations (Bongers *et al.*, 1987; Donham *et al.*, 1984). Animal's respiratory health may also be compromised by PM (Al Homidan and Robertson, 2003; Donham and Leininger, 1984). The best approach to reduce PM in and from livestock houses seems to be to prevent it from being generated. In livestock houses, PM has a high organic content, because it is mainly composed of primary coarse particles which originate from feed, manure, bedding, and animal's skin, feathers, and hair (Aarnink *et al.*, 1999; Donham *et al.*, 1986; Feddes *et al.*, 1992; Heber *et al.*, 1988; Qi *et al.*, 1992). Improved knowledge on particle morphology, primarily size, and chemical composition from livestock houses can help develop efficient and practical source-specific reduction techniques to comply with European threshold limits (Directive 1999/30 and 2008/50), and to protect the environment, and human and animal health and welfare.

Attempts to identify major sources of PM contributing to PM emissions from livestock houses have been made, although only limited data from

specific production systems related to single livestock categories are available. Comparable source contributions between and within livestock categories for different sized-particles are needed. To this end, specific methodologies which include statistical methods to calculate source contributions, standardized measuring protocols, and comprehensive field studies to characterize the morphology and composition of PM in different size fractions need to be developed. Furthermore, sampling and analysis of particulate sources are required to apportion PM to the different sources, but to date, there is lack of detailed characterization of particle size, morphology and chemical composition from sources in livestock houses. With comprehensive particle characterization and detailed source profiles, better estimates of contributions to more specific sources would be possible (Watson *et al.*, 2002). Therefore, the development of specific, accurate, and detailed source profiles for known sources from livestock houses is encouraged.

Single-particle analysis has been extensively used to characterize PM in ambient air and in other environments different from livestock houses (Adhikari *et al.*, 2003; Chen *et al.*, 2006; Conner *et al.*, 2001; Esbert *et al.*, 2001; Mamane *et al.*, 2001; Srivastava and Jain, 2007). The main advantage of single particle analysis, compared with bulk analysis, is that it can provide data from hundreds of individual particles, and therefore it can represent and describe better the properties of heterogeneous PM samples, which are comprised of a broad class of morphologies and chemical compositions. Through morphological and chemical characterization of single particles, single-particle analysis can provide an insight into origin, formation, transport, reactivity, and human health effects, as a complement to conventional bulk analysis (Chen *et al.*, 2004).

The aim of this study was to morphologically and chemically characterize individual fine and coarse PM from known sources collected from different housing systems for poultry and pigs, and to develop comprehensive morphological and chemical source profiles. More specifically, the objectives of this study were (i) to identify unique source-specific particle morphologies and define homogeneous morphological types of particles; (ii) to identify elemental source compositions and compare them amongst sources; and (iii) to determine particle size, and size distribution in each

source. The outputs of this work can provide useful information for source identification and quantification in livestock houses, improving the understanding of how PM is generated in such environments, which is essential for developing strategies for its reduction.

3.2.   Material and methods

3.2.1.   Livestock houses and source types

A total of 48 samples from known sources of PM were collected at 14 different livestock locations in The Netherlands, including seven different housing systems for poultry and pigs. Two farms were sampled for each livestock housing system. Table 3.1 describes surveyed livestock houses, livestock species, type of housing system, ventilation system, number of animals, animal age, and collected PM sources at each farm. All surveyed livestock houses used automatically distributed feeding systems with crumbles or pelleted feed. The collected PM source types were chosen at each farm depending on the housing system. All farms were sampled for manure and concentrate feed. Feathers were collected in all poultry houses, and hair in all pig houses. We also collected wood shavings used as bedding material in broiler and turkey houses; and skin in pig houses, but only from sows, because it was impractical to collect such source from younger animals (piglets and growing-finishing pigs).

3.2.2.   Known source sample collection and preparation

Sampling was conducted during morning (from 09:00 to 12:00) at each livestock house. A representative sample from each PM source was obtained by randomly sampling different spots in the livestock house. A total of 200 to 500 grams of feed, clean bedding, and fresh manure samples were collected at each location. Fresh poultry excreta were collected directly from the floor or house surfaces. Pig feces were randomly sampled in each livestock house from slatted or concrete floor. A total of 10 to 50 grams of hair, feathers, and skin, were directly collected from clean animals. Samples were stored in clean sealable polyethylene bags, and transported to the laboratory and stored under refrigeration. Each sample was then mixed to achieve a uniform sample and the samples were dried in the oven for 12 h at

70°C. Dried samples were crushed in a ball mill during 1.5 minute at 250 rpm. Dried and milled samples were stored at room temperature (20-25°C).

Table 3.1. Description of surveyed livestock houses and collected PM sources.

Livestock species	Housing system	Farm location	Ventilation	Number of animals	Age (weeks)	Collected PM source types
Poultry	Broilers - bedding	1	Tunnel	50,400	4	Fresh excreta
		2	Roof	2675	3	Feed (crumbles and pellets)
	Turkeys - bedding	1	Ridge	5000	12	Feathers
		2	Ridge	4040	10	Wood shavings
	Laying hens - floor	1	Tunnel	3850	71	Fresh excreta
		2	Tunnel	16,500	22	Feed (crumbles and pellets)
	Laying hens - aviary	1	Tunnel	24,712	71	Feathers
		2	Tunnel	35,000	50	
Pigs	Piglets- slatted floor	1	Roof	125	8	
		2	Roof	75	9	Fresh feces
	Growing-finishing pigs - partially slatted floor	1	Roof	120	16	Feed (pellets)
		2	Roof	60	20	Hair
	Dry and pregnant sows - group housing	1	Roof	39	-	Fresh feces
		2	Roof	46	-	Feed (pellets) Hair Skin

A representative sample of ambient outdoor fine and coarse PM at each location was also collected on each sampling day. These PM samples were collected using a virtual cascade impactor (RespiCon, Wetzlar, Germany). This impactor simultaneously sampled PM2.5 and PM10-2.5 particles. A portable pump (Genie VSS5, Buck Inc, U.S.) was used to suck air through the impactor at a constant flow of 3.11 L min^{-1}. Particles were collected on polycarbonate filters (37 mm Ø, 5 µm pore size), and stored before analysis. Sampling time varied from 30 to 60 minutes, aiming at particle loads appropriate for single-particle SEM analysis of 5 to 20 µg particles cm^{-2} filter (Willis *et al.*, 2002).

3.2.3. Size-segregated PM generation and measurements

To obtain size-segregated PM samples from the different known sources, a mechanical agitation system was used. Each milled source was agitated in a customized laboratory stainless steel dust generator (Figure 3.1). Because the dust potential of the different sources was not the same, and aiming at particle loads appropriate for single-particle SEM analysis of 5 to 20 µg particles cm^{-2} filter (Willis *et al.*, 2002), we varied and adjusted the amount of sample and the dust generation time. Approximately 0.2 grams of milled feathers and skin, 2 to 3 grams of milled manure, hair and wood shavings, and 40 grams of milled feed were used in the dust generator, agitated at 200 rpm. Sampling time varied from 1 minute (feathers), 2 minutes (manure), 4 minutes (skin), 20 minutes (hair), 3 hours (wood shavings), and 7 hours (feed). The PM2.5 and PM10-2.5 generated particles during agitation were collected using a virtual cascade impactor (RespiCon, Wetzlar, Germany) and a portable pump, using polycarbonate filters. Loaded filter samples were stored in sealed filter cassettes at room temperature (20-25ºC) before analysis.

At the same time, an optical particle counter (OPC, model 1.109, Grimm Aerosol Technik GmbH & Co., Ainring, Germany) was used during the generation process to monitor particle-size distribution (PSD) per source. The inlet of the device was connected to the dust generation chamber. Air was sampled through the inlet at 1.2 L min^{-1}. The optical particle counter sampled and counted particles in 31 size ranges, from 0.25 to 32 µm in diameter using light scattering principle. Recorded values were stored every 6 seconds. Sampling time was 7 minutes per sample. This instrument was also used to determine PSD of outdoor particles, outside farm locations.
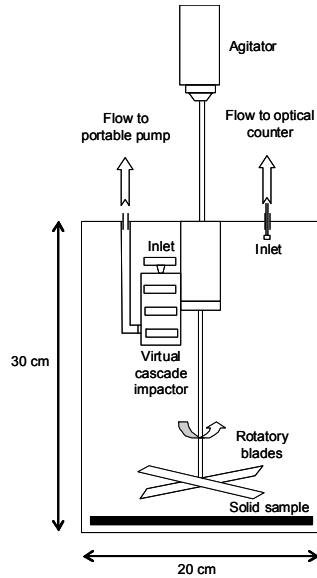
Figure 3.1. Schematic layout of dust generation process, measurements and position.

3.2.4. Scanning electron microscopy analysis

Generated particles collected on polycarbonate filters were analyzed using high-resolution scanning electron microscopy (SEM) (JEOL, JSM-5410) combined with energy-dispersive X-ray analysis (EDX) (Link Tetra Oxford Analyzer). The SEM-EDX system had a thin window EDX detector enabling X-ray detection of elements with atomic number ≥ 6 (carbon). With SEM-EDX, individual particles could be imaged by SEM while EDX provided information on the chemistry of individual particles. The interaction between the electron beam produced by the SEM, and the atoms in the sample results in emission of X-rays, whose energies (keV) are characteristic of specific elements present in the sample (Willis *et al.*, 2002).

A small section (approximately 1 cm²) of the loaded polycarbonate filter from fine and coarse fractions was cut and mounted on a 12 mm carbon stub with a double-sided carbon adhesive tape. Each sample was then coated with carbon using a vacuum evaporator to create a conductive coating for exposure to the SEM electron beam.

The SEM-EDX was conducted manually, operated under the same conditions throughout the study: accelerating voltage 10 keV, working

distance 15 mm, electron probe current of 3 nA, magnifications 1000x for coarse PM, and 1800x for fine PM, and X-ray acquisition time 60 s per particle. Secondary electron mode was used for particle location, measurement, analysis, and image acquisition.

Uniformity of particle deposition on the filter was verified examining the filter prior to analysis at low magnification (300x). Then, at least three fields of view (spots) per filter sample were analyzed. On each analyzed field, both an image (photomicrograph at 1000x or 1800x) and single particle X-ray spectra of every particle found in that field were obtained and stored. Within each field, the minimum projected area diameter for the coarse particles was set at 1 μm. The minimum projected area diameter for the fine particles was set at 0.1 μm (Conner *et al.*, 2001). These limits were set because otherwise the detection and analysis of smaller particles was not reliable at the used magnifications. A total of 25 to 50 individual particles were analyzed in each sample. All spectra were confirmed and checked manually to correct for the contribution of the filter material (C and O). As samples were not flat but comprised complex sized and shaped particles, the elemental analysis was used in a relative semi-quantitative way (Kasparian *et al.*, 1998). Elemental concentrations were normalized to 100% (Sitzmann *et al.*, 1999).

Photomicrographs of each field of view were acquired at normal gray and saved in tif format (1024x768 resolution). These images which included all analyzed particles were further analyzed using the Object Based Image Analysis (OBIA) approach (Blaschke, 2010) using FETEX 2.0 Software (Ruiz *et al.*, 2010). This image analysis and processing system automatically detected each particle object and calculated the particle projected area. From the particle area, the projected area diameter (D_p) was calculated, defined as the diameter of a perfect circle fitted to the measured area of the particle (equation 1). This diameter measured by microscopy is also referred to as the physical or geometric diameter (Conner *et al.*, 2001).

$$D_p = 2 \times \sqrt{\frac{Area}{\pi}} \qquad\qquad (1)$$

3.2.5. Data analyses

Particle types and morphologies were qualitatively analyzed based on the SEM images. Different types of particles were identified in each SEM field of view. These particle types were morphologically described in terms of shape (rounded, spherical, fibrous, flake, angular, aggregate, irregular, flattened, long-thin), surface (layered, smoothed, cracked), edges and borders (sharpness), texture (smooth, grape-like, and rough), and opacity, amongst others (McCrone, 1992; NIST, 2010). In this way, different types of particles were determined in each source, in fine and coarse PM. More than 300 images were qualitatively analyzed.

Particle chemical compositions were summarized to obtain the average relative elemental concentrations per source in fine and coarse PM, pooled by livestock category. The relative elemental composition of the PM in the different sources and in each fraction was compared using analysis of variance with SAS software (SAS, 2001). To test multivariate differences between sources, and identify which elements (variables) discriminated best amongst sources per fraction, we performed a stepwise discriminant analysis using SAS software (SAS, 2001). With this analysis, variables were chosen according to the significance level of an F-test from an analysis of covariance, where the variables already chosen act as covariates and the variable under consideration act as dependent variables.

Data on size were summarized to obtain the average D_p per source in fine and coarse PM, pooled by livestock category. The average D_p of the PM in the different sources and in each fraction was compared using analysis of variance with SAS software (SAS, 2001).

To determine the PSD per source, we calculated the standardized number fraction (Δf_i) from the frequency of particles (F_i) within a size range (Δd_i) in each source. The standardized number fraction of particles for the ith size range was calculated with equation 2:

$$\Delta f_i = \frac{\left(\dfrac{F_i}{\Delta d_i}\right)}{N} \qquad (2)$$

where:

Δf_i= Standardized number fraction in units of μm^{-1} for the ith size range

F_i= Frequency (number) of particles within a size range

Δd_i= Particle size range, calculated as the difference between the upper and lower limit of the sampling interval (size range measured by the instrument) within each group of particles

N= Total number of particles measured by the instrument (sum of all size ranges).

We also calculated the standardized mass fraction by multiplying the particle number concentrations by an estimated particle mass per source, assuming all particles were spherical, and assuming a value for particle density. Density values of 1.2 g cm^{-3} (feathers), 2.6 g cm^{-3} (feed), 1.3 g cm^{-3} (hair), 1.5 g cm^{-3} (manure and wood shavings), 1.4 g cm^{-3} (skin), and 2.1 g cm^{-3} (outside) were used (McCrone, 1992). The calculation of mass from numbers was done following equation 3:

$$m_i = n_i \times \rho_p \times v_{pi} = n_i \times \rho_p \times \left[\frac{4}{3} \times \pi \times r_i^3 \right] = n_i \times \frac{\rho_p \times \pi \times \left(d_{g_i} \right)^3}{6} \qquad (3)$$

where:

m_i= particle mass for the ith size range of particles

n_i= number of particles measured by the instrument for the ith size range

ρ_p= particle density per source

v_{pi}= particle spherical volume for the ith size range

r_i= equivalent radius of a spherical particle for the ith size range

d_{gi}= mean geometric particle diameter measured by the instrument in the ith size range

This size distribution was also standardized and divided by the total mass of particles to obtain the standardized mass fraction in the same way as for standardized number fraction (equation 2).

3.3. Results

3.3.1. Particle types and morphology (fine and coarse)

Particle morphologies were qualitatively analyzed in each fraction based on SEM images. Different types of particles were identified and thoroughly described for each source. Scanning electron microscopy images of particles collected on polycarbonate filters are shown below.

3.1.1. Feathers

Feathers showed a mixture of irregular, mostly flattened particles in fine and coarse PM. Three morphological types were identified: soft and "fluffy" particles, sometimes bent (Figure 3.2a and b); rounded, flake-like flattened, sometimes aggregate particles with rough texture (Figure 3.2c and d); and stiff, elongated, and pointed particles (Figure 3.2e and f). Each type generally coincided with different livestock categories. In broilers, small soft and "fluffy" particles were dominant in fine and coarse PM. In laying hens, besides showing some soft and "fluffy" structures, also flake-like flattened particles and elongated particles were dominant in fine and coarse PM. Turkeys showed mostly soft and "fluffy" particles in the fine fraction (Figure 3.2g); whereas flake-like flattened and elongated particles were also abundant in coarse PM (Figure 3.2h).

(a)   (b)
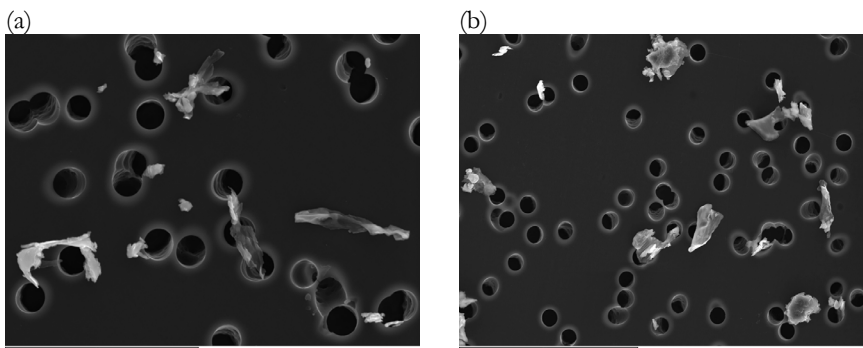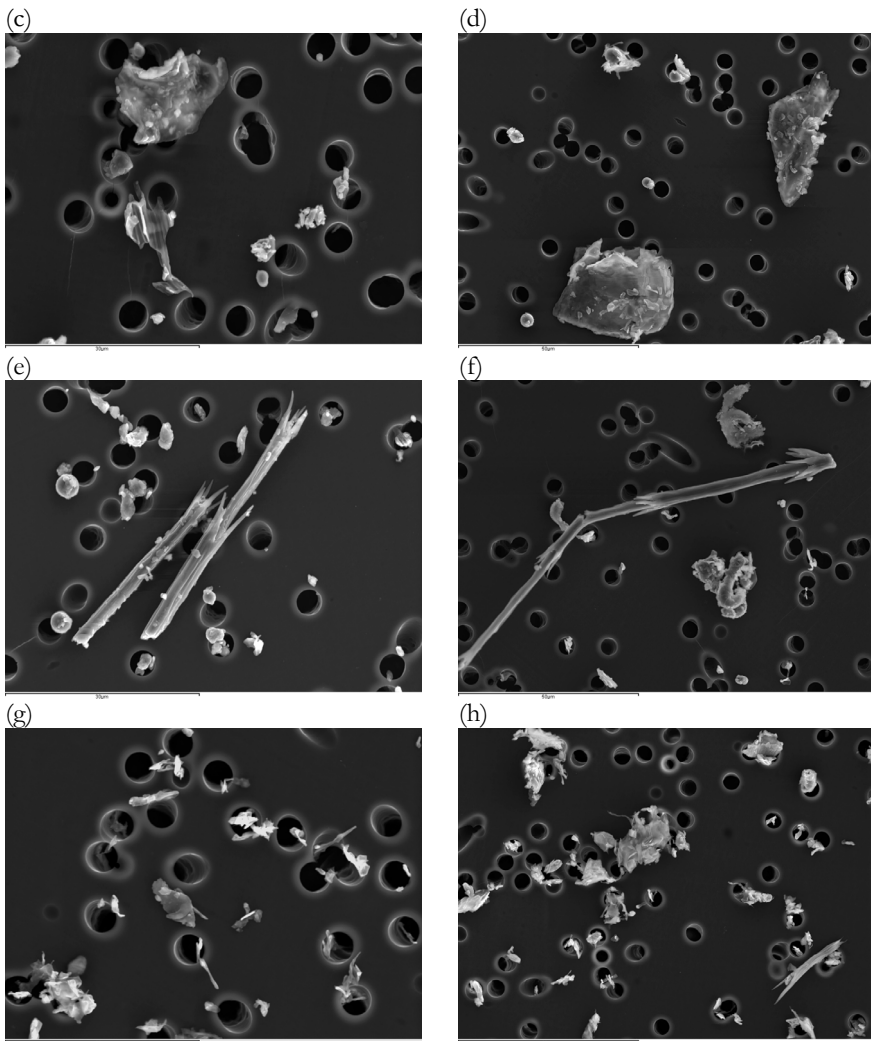
Figure 3.2. Particles from feathers. (a) Long and "fluffy" particles from broilers in fine PM. (b) Mixture of "fluffy" particles showing different silhouettes from broilers coarse PM. (c) Big rounded, flattened particle together with smaller "fluffy" particles from laying hens in floor system fine PM. (d) Rounded and triangular flattened particles from laying hens in floor system coarse PM. (e and f) Stiff, elongated, and pointed particles from laying hens in aviary system fine PM (e) and coarse PM (f). (g) Soft and "fluffy" particles from turkeys in fine PM. (g) Mixture of "fluffy", flake-like, and elongated particles from turkeys in coarse PM. Images on the left: fine PM, scale bar 30 µm. Images on the right: coarse PM, scale bar 50 µm. Note 5 µm diameter filter pores, shown as round dark holes.

3.1.2. Feed

Four general morphological types of feed particles were identified: rounded and deposited particles, sometimes fragmented (mainly seen in broilers and turkeys) (Figure 3.3a and b); geometric quadrangular, cubic (Figure 3.3c and d) or bar-shaped particles (Figure 3.3e and f); and angular, cracked, fragmented particles (Figure 3.3g and h). All types were randomly found in fine and coarse PM amongst all livestock categories.

(g) (h)
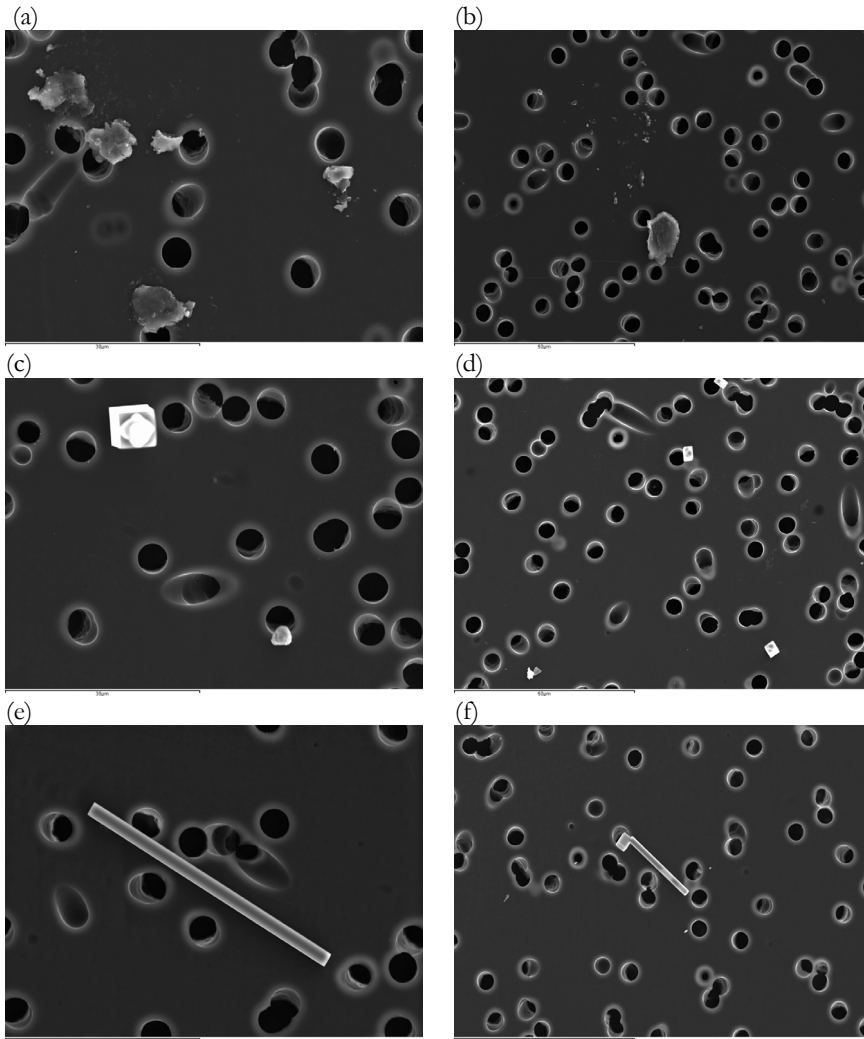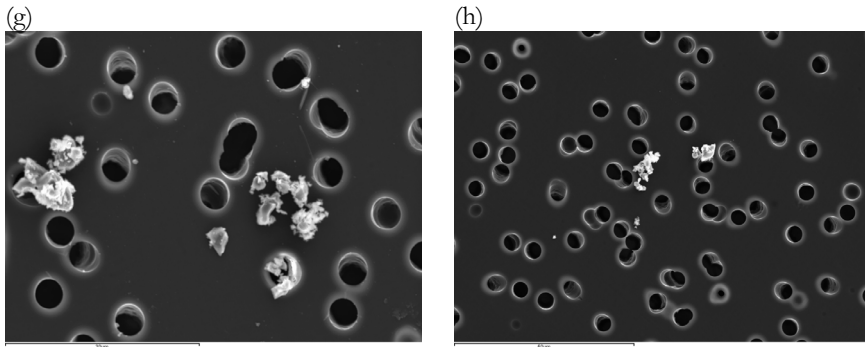
Figure 3.3. Particles from feed. (a and b) Rounded and flattened, smooth particles from broilers fine PM (a) and also rests of fragmented particles in coarse PM (b). (c and d) Cubic bright particles from laying hens aviary system fine PM (d) and from sows coarse PM (d). (e and f) Single bar-shaped particles from sows fine PM (e) and laying hens floor system coarse PM (f). (g and h) Several angular, cracked, fragmented particles from laying hens aviary fine PM (g) and growing-finishing pigs coarse PM (h). Images on the left: fine PM, scale bar 30 μm. Images on the right: coarse PM, scale bar 50 μm. Note 5 μm diameter filter pores, shown as round dark holes.

3.1.3. Hair

Pig's hair showed long-thin particles. Two types of hair particles were identified in fine and coarse PM: thin pointed particles (Figure 3.4a and b); and striated tubular particles (Figure 3.4c and d).

(a) (b)
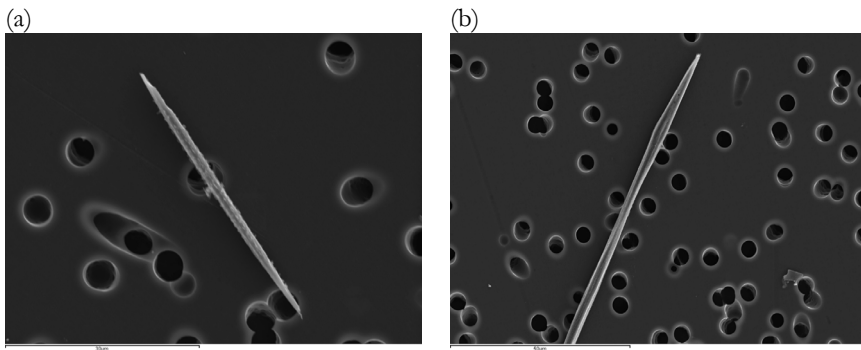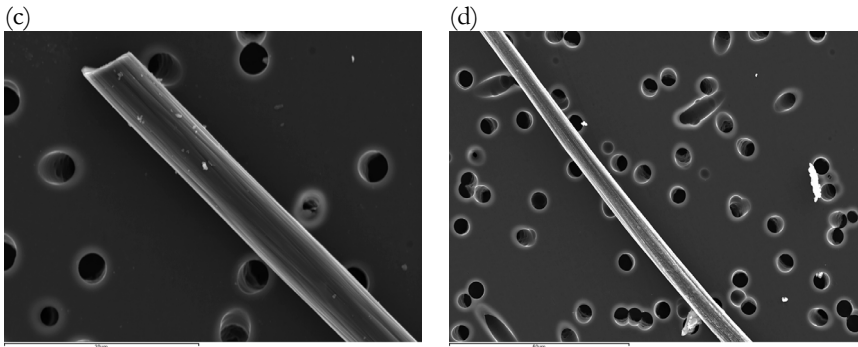
Figure 3.4. Particles from hair. (a and b) Long-thin pointed particles from growing-finishing pigs fine PM (a) and from piglets coarse PM (b). (c and d) Thick and striated tubular particles from growing-finishing pigs fine PM (c) and from sows coarse PM (d). Images on the left: fine PM, scale bar 30 μm. Images on the right: coarse PM, scale bar 50 μm. Note 5 μm diameter filter pores, shown as round dark holes.

3.1.4. Manure

Manure particles showed two morphological types: rounded, spherical, and smooth particles; and fragmented, rough, and angular particles. Rounded spheres were only identified in poultry excreta, in fine and coarse PM. Apart from rounded spheres being more abundant in poultry excreta, irregular, angular particles were also identified in poultry manure. Rounded spheres were sometimes present as individual particles (Figure 3.5a), and agglomerated with fragmented angular particles (Figure 3.5b), or highly agglomerated forming grape-like structures (Figure 3.5c and d). Rough and ciliated rounded spheres were identified in turkeys and laying hens manure (Figure 3.5e and f). Fragmented, layered, angular particles were the dominant particles in pigs in fine (Figure 3.6a and b) and coarse PM (Figure 3.6c and d).
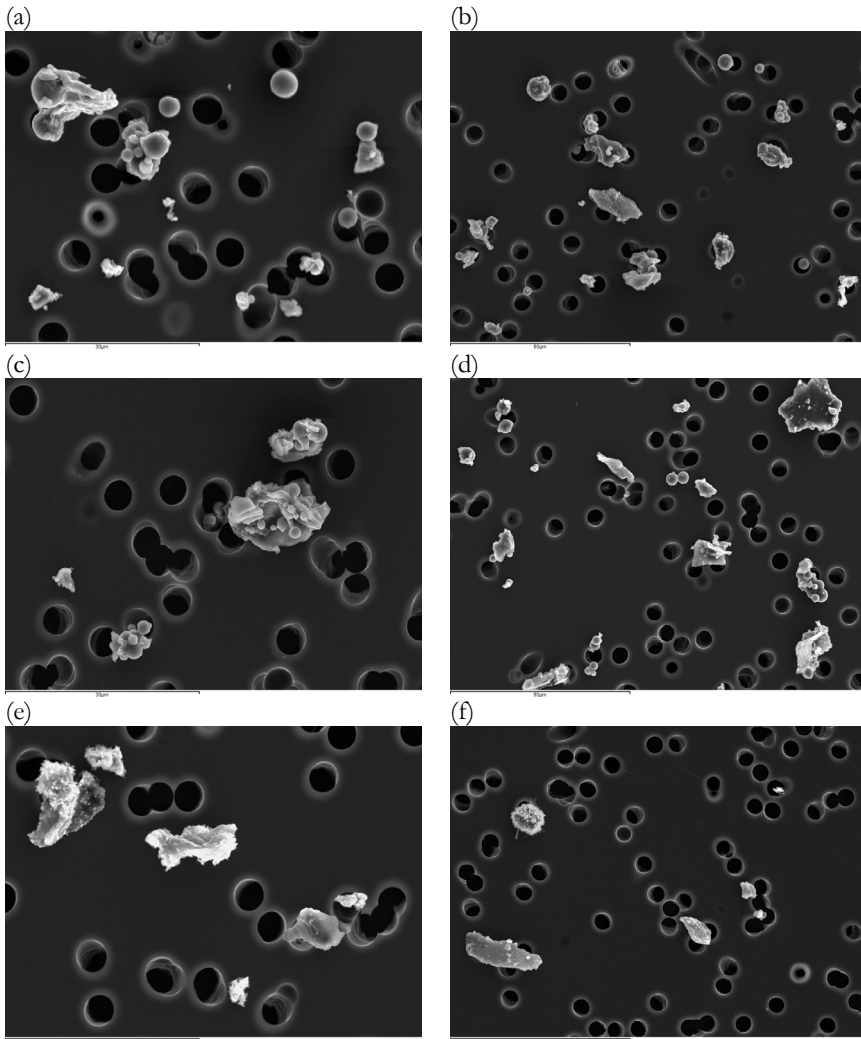
Figure 3.5. Manure particles from poultry. (a) Mixture of single rounded spherical and irregular particles from laying hens aviary system fine PM. (b) Few single rounded spherical and more abundant fragmented angular particles from laying hens aviary system coarse PM. (c) Agglomerated grape-like particles from broilers fine PM. (d) Some grape-like agglomerated particles and fragmented angular particles from turkeys coarse PM. (e and f) Mixture of rough, fragmented, angular and ciliated rounded particles from turkeys fine PM (f) and from laying hens floor system coarse PM (f). Images on the left: fine PM, scale bar 30 μm. Images on the right: coarse PM, scale bar 50 μm. Note 5 μm diameter filter pores, shown as round dark holes.
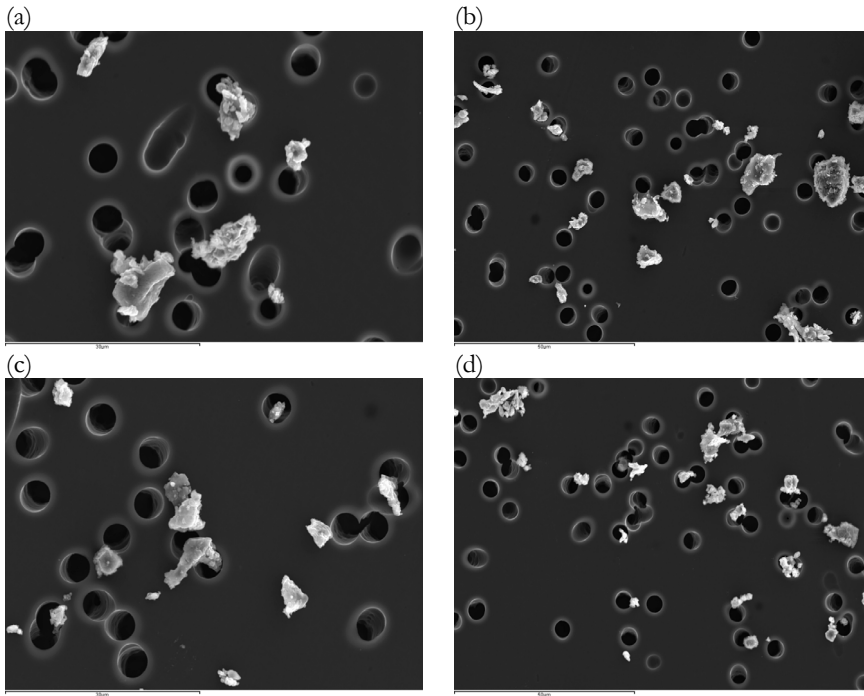
Figure 3.6. Manure particles from pigs. (a) Fragmented angular particles from piglets fine PM. (b and c) Mixture of fragmented, layered, angular and more rounded particles from growing-finishing coarse PM (b) and from growing-finishing fine PM (c). (d) Abundant layered and angular particles from sows coarse PM. Images on the left: fine PM, scale bar 30 μm. Images on the right: coarse PM, scale bar 50 μm. Note 5 μm diameter filter pores, shown as round dark holes.

3.1.5. Skin

Sow's skin particles were morphologically homogeneous and showed a single type, as big, rounded, thin, flattened, flake-like, transparent particles in fine (Figure 3.7a and c) and coarse PM (Figure 3.7b and d). These flake-like particles presented a smooth surface (Figure 3.7a and c), although some of them presented rough surfaces caused by deposited particles on top (Figure 3.7b and d).
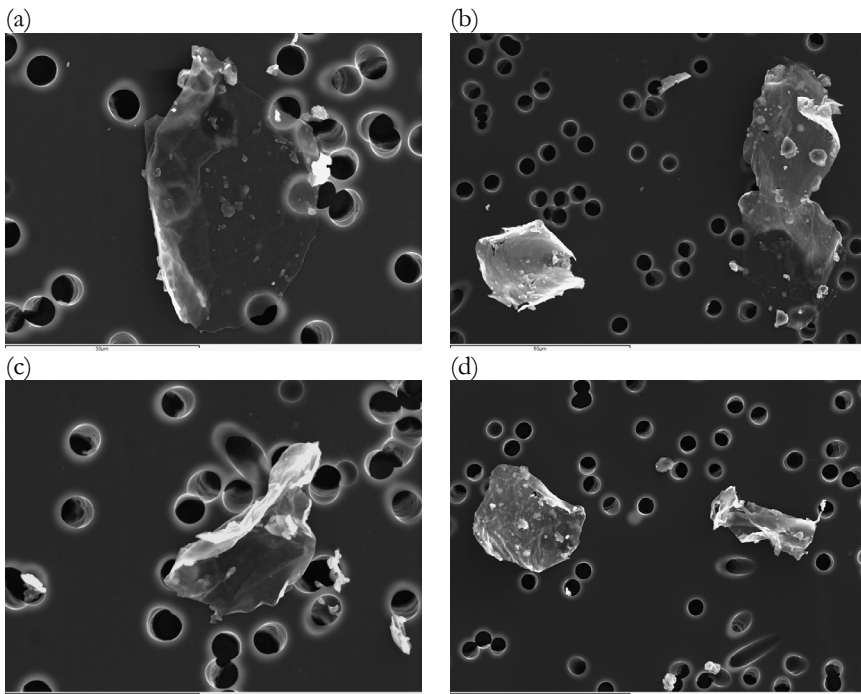
Figure 3.7. Particles from skin. All particles from sows. (a) Big, transparent, smooth, and flat particle in fine PM. (b) Rounded flake-like particles from coarse PM. (c) Folded and thin particle from fine PM. (d) Rough surfaces caused by deposited particles on top of flattened particles from coarse PM. Images on the left: fine PM, scale bar 30 μm. Images on the right: coarse PM, scale bar 50 μm. Note 5 μm diameter filter pores, shown as round dark holes.

3.1.6. Wood shavings

Wood shaving particles showed two types of particles: flattened, round with irregular borders, others elongated and bent in fine PM (Figure 3.8a and c); and mostly fibrous particles with sharp edges identified in coarse PM (Figure 3.8b and d).
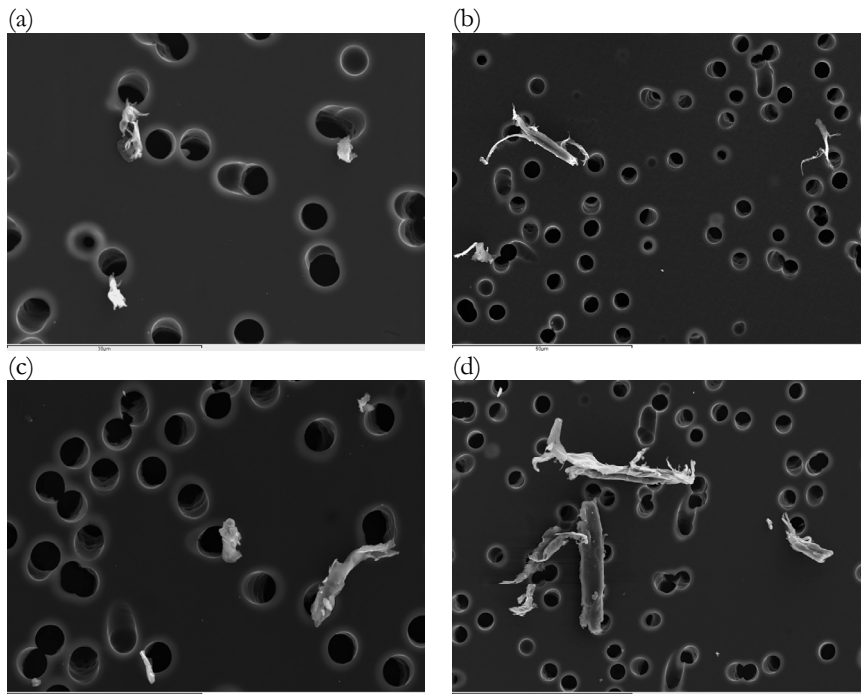
Figure 3.8. Particles from wood shavings. (a) Rounded flattened particles from broilers fine PM. (b) Fibers from broilers in coarse PM. (c) Rounded and elongated, bent particle from turkeys fine PM. (d) Fibrous particles with very sharp edges from broilers in coarse PM. Images on the left: fine PM, scale bar 30 μm. Images on the right: coarse PM, scale bar 50 μm. Note 5 μm diameter filter pores, shown as round dark holes.

3.1.7. Outside source

Particles from outside farm sources showed heterogeneous morphologies. Dominant particles were generally small, irregular angular, cracked fragmented particles (sometimes aggregate) (Figure 3.9a and b); and geometric quadrangular, bar-shaped or cubic particles (Figure 3.9c and d).
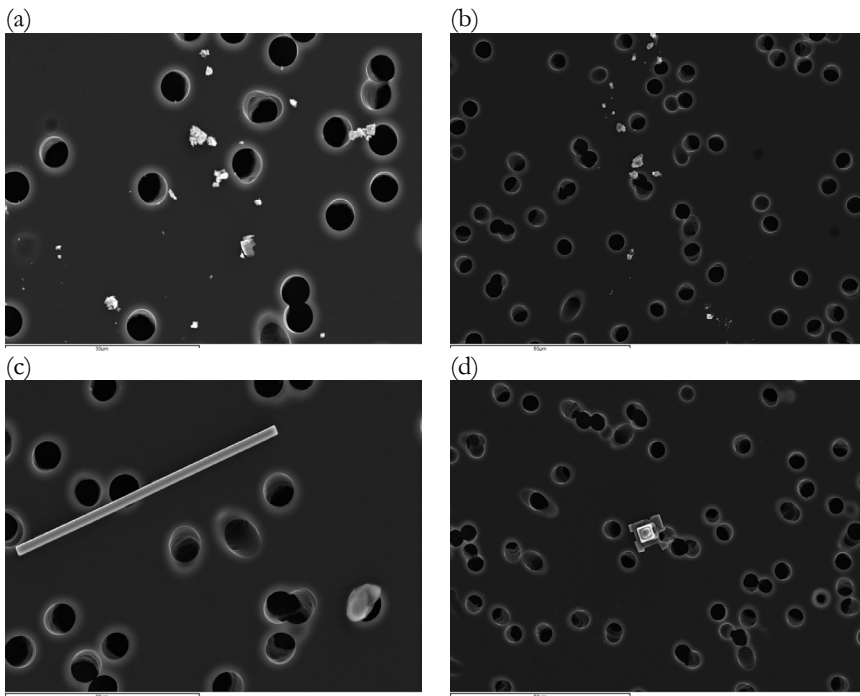
Figure 3.9. Particles from outside livestock houses. (a and b) Irregular angular, cracked, and fragmented particles in fine PM (a) and coarse PM (b). (c) Bar-shaped particle in fine PM. (d) Cubic particle in coarse PM. Images on the right: coarse PM, scale bar 50 μm. Note 5 μm diameter filter pores, shown as round dark holes.

3.3.2. Chemical composition (fine and coarse)

Average relative elemental concentrations were calculated per source in fine and coarse PM, pooled by livestock category. Figure 3.10 (fine PM) and Figure 3.11 (coarse PM) present average element relative concentration (expressed in percentage) for particles from each source, together with significant differences in average values of element concentrations amongst sources. Hair was not included in the analysis because it showed very high carbon and oxygen peak in the SEM-EDX which was confounded with the background filter composition. Similar elements were identified in fine and coarse PM, as well as in different sources. Presence of N, Na, Mg, Al, Si, P, S, Cl, K, and Ca were identified in all sources, in fine and coarse PM. Generally, differences in these elements amongst sources were obtained between feed, outside, wood, skin, and the rest of sources; or between manure and the rest of sources. Traces of heavy elements (metals), with

atomic numbers greater than 20 (such as Fe, Ni, Cu, Zn, Ag, Pb, Sn, Ba, and Cu) were mainly identified in feed and outside, and to a smaller extent in wood shavings. Feathers showed presence of Fe, Ni, Sn, and Cr, in relative elemental concentrations below 0.6%. Manure showed presence of Fe, Cu, and Sn in relative elemental concentrations also below 0.6%, but 6% of Au. Other elements not shown in Figure 3.10 and Figure 3.11, were detected in some particles in fine and coarse PM (Co in feed, manure, and outside), and others only in coarse PM (Br, Ti, V, and Sb in feed, wood shavings, and outside), in relative concentrations below 0.2%, and showing no statistical significant differences amongst sources.

Similar trends in elemental levels were detected amongst sources and amongst fine and coarse PM. Five peaks coinciding with high relative levels of N, Na, S, Cl, and Ca could be seen in all sources. Manure showed the highest relative levels of N, Mg, P, and K compared with the rest of sources; whereas feathers, besides also peaking at N, showed lower relative levels of P and Mg than manure, and higher S and Cl. Skin showed the highest S levels compared with the rest of the sources. Wood shavings showed the highest levels of Cl and Na. Feed showed the highest levels of Si and Ca. Outside particles showed the highest levels of Al, in fine PM.

Results from the discriminant analysis confirmed the differences in relative elemental concentrations amongst sources, presented in Figure 3.10 and Figure 3.11. The first five common variables that best discriminated amongst different sources were P, N, Cl, S, and K. Table 3.2 and Table 3.3 show the summary of the stepwise discriminant analysis for each variable considered, showing the squared partial correlation, the F-statistic, and the probability level, from the one-way analysis of covariance. In fine PM, order of entrance into the discriminant process was: P, N, Cl, S, K, Si, Na, Al, Ca, Mg, Sn, and Pb (Table 3.2). In coarse PM, the order of entrance into the discriminant process was: P, N, K, S, Cl, Al, Ca, Cr, Na, Mg, Ba, and Fe (Table 3.3).
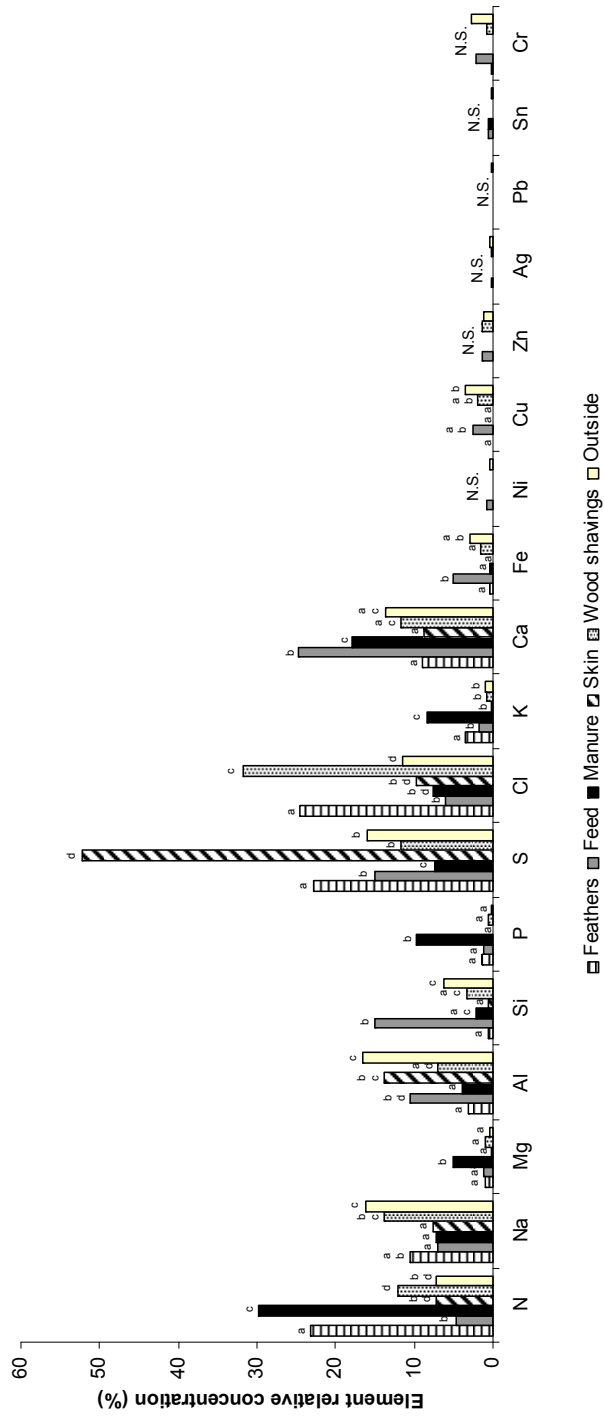
Figure 3.10. Average element relative concentration (%) for particles from different sources in fine PM2.5. Averages within an element lacking common superscript letter are significantly different (P < 0.05). (N.S. stands for non significant differences).
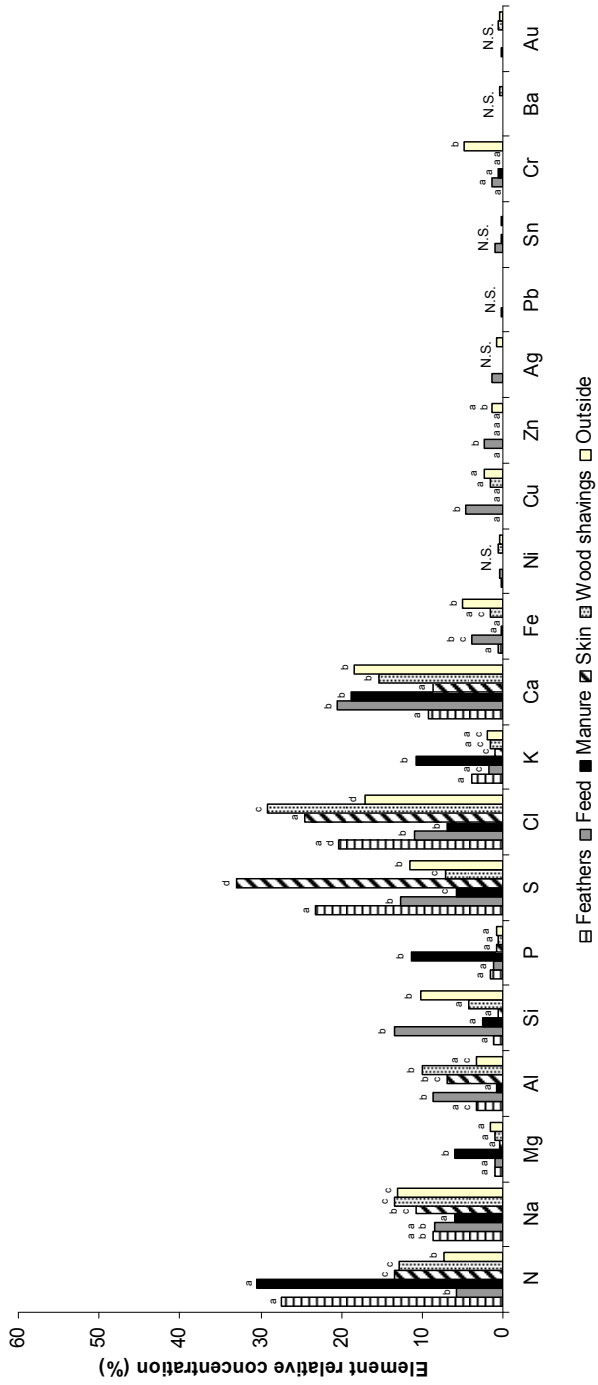
Figure 3.11. Average element relative concentration (%) for particles from different sources in coarse PM10-2.5. Averages within an element lacking common superscript letter are significantly different (P < 0.05). (N.S. stands for non significant differences).

Table 3.2. Summary of the stepwise discriminant analysis showing the squared partial correlation (Partial R-Square), the F-statistic (F-value), and the probability level (Pr > F), from the one-way analysis of covariance in fine PM.

Order of entrance in the model	Element	Partial R-Square	F-value	Pr > F
1	P	0.2576	113.04	< 0.0001
2	N	0.2446	105.43	< 0.0001
3	Cl	0.2392	102.31	< 0.0001
4	S	0.1456	55.41	< 0.0001
5	K	0.1161	42.67	< 0.0001
6	Si	0.0475	16.2	< 0.0001
7	Na	0.0406	13.74	< 0.0001
8	Al	0.0318	10.64	< 0.0001
9	Ca	0.0151	4.96	0.0002
10	Mg	0.0125	4.09	0.0011
11	Sn	0.0083	2.71	0.0190
12	Pb	0.0057	1.85	0.0997

Table 3.3. Summary of the stepwise discriminant analysis showing the squared partial correlation (Partial R-Square), the F-statistic (F-value), and the probability level (Pr > F), from the one-way analysis of covariance in coarse PM.

Order of entrance in the model	Element	Partial R-Square	F-value	Pr > F
1	P	0.2963	139.97	< 0.0001
2	N	0.2629	118.46	< 0.0001
3	K	0.1772	71.51	< 0.0001
4	S	0.1371	52.72	< 0.0001
5	Cl	0.1181	44.43	< 0.0001
6	Al	0.0372	12.82	< 0.0001
7	Ca	0.0208	7.04	< 0.0001
8	Cr	0.0137	4.59	0.0004
9	Na	0.0132	4.42	0.0005
10	Mg	0.0108	3.61	0.0030
11	Ba	0.0073	2.42	0.0340
12	Fe	0.0071	2.37	0.0375

3.3.3. Size and size distributions

In each source, particle size, expressed as D_p, was determined from SEM images using image analysis software. Particle-size distribution was determined by the light scattering principle during aerosolization in the dust generator.

3.3.1. Particle size

For all sources (except for hair) average D_p in fine PM was from 35 to 46% lower ($P < 0.005$) compared with coarse PM. Skin and hair showed the largest particle sizes (D_p equal to 13 μm in fine PM, and 18 μm in coarse PM); whereas feed and outside particles showed the lowest sizes (D_p equal to 2 μm in fine PM, and 3 μm in coarse PM). Average D_p (standard deviation, SD) for the different sources in fine and coarse PM are shown in Table 3.4.

Table 3.4. Average estimated projected area diameter (D_p) from particle areas from SEM images and standard deviation (SD), for different sources in fine and coarse PM fractions (N.S. stands for non significant differences).

Source	n	Fraction	Average	SD	P-value
Feathers	398	PM2.5	3.9	2.9	< 0.0001
	431	PM10-2.5	5.6	5.4	
Feed	416	PM2.5	2.1	2.2	< 0.0001
	405	PM10-2.5	3.0	2.7	
Hair	34	PM2.5	11.7	5.2	N.S.
	36	PM10-2.5	10.8	5.8	
Manure	644	PM2.5	4.0	2.3	< 0.0001
	942	PM10-2.5	5.5	2.8	
Skin	27	PM2.5	13.4	8.0	< 0.05
	42	PM10-2.5	18.1	8.0	
Wood shavings	130	PM2.5	4.1	3.3	< 0.0001
	212	PM10-2.5	5.9	5.2	
Outside	350	PM2.5	2.1	1.9	< 0.0001
	246	PM10-2.5	3.0	2.9	

3.3.2. Particle-size distribution

Figure 3.12 shows measured average particle number-size distribution for each source in log-scale, calculated from the average number of particles per size range for each source. All sources showed the highest number of particles in the lowest size ranges and the lowest number of particles in the highest size ranges. Particles in the size range from 0.25 to 0.28 μm were the most abundant in all sources, being this the minimum size range measured by the instrument. From approximately 0.6 μm, differences amongst size distributions from sources became evident. From this size range onwards, two different size distributions were observed: size distribution from feed

and outside which decreased more or less linearly; and size distribution from the rest of sources which showed a less sharp decrease. For the rest of sources, two peaks, one at 0.8 to 0.9 μm, and another at 4 to 5 μm, could be identified, after which particle numbers decreased linearly. All sources showed a peak in the last size range (particles bigger than 32 μm), showing a relatively high number of very big particles present in all source.
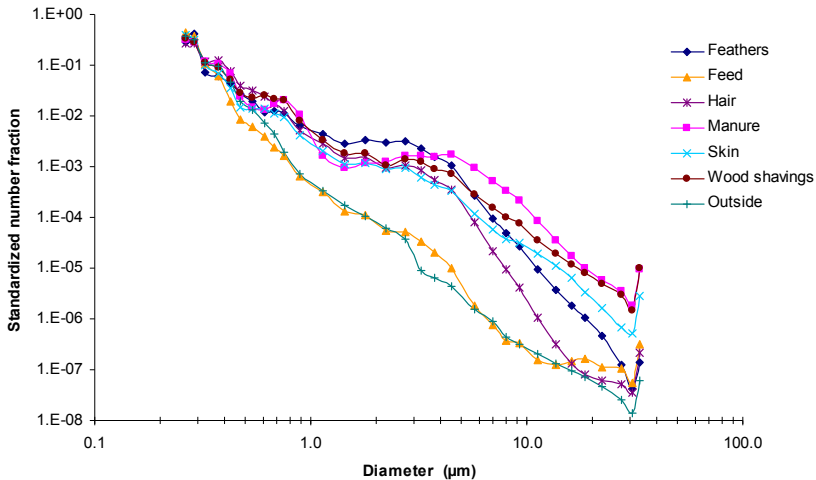
Figure 3.12. Standardized number fraction size distribution for particles from different sources (log-scale).

Figure 3.13 shows the average particle mass-size distributions for each source in log-scale, calculated from the average mass of particles per size range for each source. Particle mass-size distributions showed high masses in the lowest size ranges, in the middle size ranges, but also in the highest size ranges. High mass for feed and outside was observed in the minimum size range measured by the instrument (size range from 0.25 to 0.28 μm). High masses were observed in the small diameters especially for outside source. For the rest of sources, high masses were found at 4 to 5 μm, where feed and outside showed their minimum mass. Above 5 μm, the mass of feathers and hair decreased more sharply, showing lower masses compared with manure, skin, and wood shavings. Above 5 μm, feed and outside masses increased. Manure's mass distribution showed four very clear peaks

at 0.25, 0.4, 0.8, and 4 μm. Again, all sources showed a peak in the last size range, corresponding to particles bigger than 32 μm.
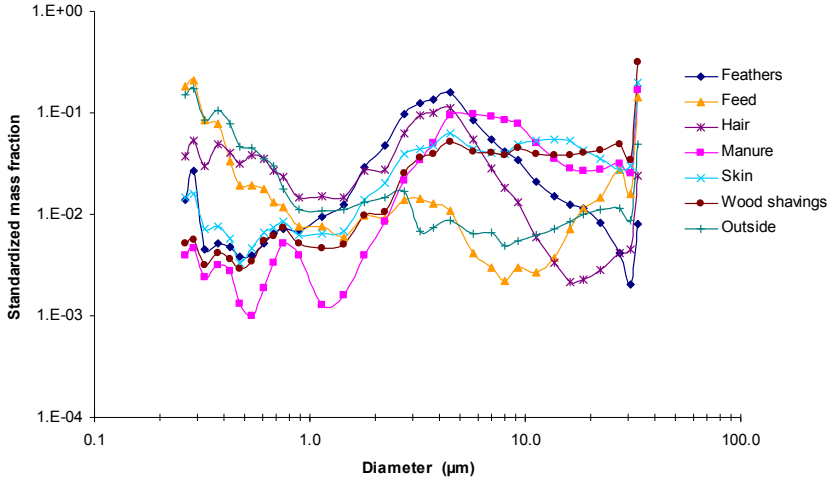
Figure 3.13. Standardized mass fraction size distribution for particles from different sources (log-scale).

3.4. Discussion

The application of SEM-EDX to individual particles from collected sources in different livestock housing systems for poultry and pigs demonstrated that sources of PM differed in particle morphology, elemental composition, and size. This study gives a detailed and complete analysis of potential sources of PM from livestock houses including different housing systems for poultry and pigs in size-segregated PM. Qualitative results revealed different particle morphological types and unique morphological features related to each source. Some of the identified particle types coincided and could be related to a specific livestock category (e.g. type of feathers and manure), although others were generally randomly found in all livestock categories (e.g. types of feed particles). The use of digital image analysis software could be useful to extract morphological characteristics and quantify further differences.

The different morphological types of particles identified in the SEM analysis could be partly explained by the different livestock production systems.

Particle types from feathers could be explained by the feather structure and development process, related to different poultry production systems. All types of particles from feathers identified in the SEM corresponded to the lighter parts, probably more prone to become airborne than other heavier parts of the feather. In broiler houses, younger birds than in laying hen houses can be found, because one-day-old birds are introduced in the broiler houses, and are slaughtered only after 5 to 7 weeks of age. In our study, farms with 3 to 4 week-old broilers were sampled and broiler's feathers were seen as fine feathers (plumules or down feathers), with shorter shafts than adult feathers, with "fluffy" structure to provide a high level of insulation to young birds, thus easily airborne. In laying hen houses, hens older than 20 weeks are generally found. Therefore, laying hen's feathers have more mass than and differ from down feathers. Laying hen's feathers and also turkey's feathers were more similar to contour feathers than to down feathers. Contour feathers consist of a shaft onto which a feather vane is attached (Leeson and Walsh, 2004). Adult and juvenile birds, however, can also show down feathers placed beneath contour feathers, as also identified in laying hens. The feather vane, moreover, is composed of filaments, called barbs, which have rows of interlocking barbules or hooklets that give the feather its shape and rigidity (Leeson and Walsh, 2004). Barbules (named hooklets after their pointed structure) are also fine structures, easily airborne, which were abundant in samples from laying hens feathers, and clearly identifiable by their pointed and elongated morphology.

The existence of two very distinctive morphological types of manure particles between poultry and pigs could be explained by the particular poultry excretory system, where urea is converted chemically to uric acid. Birds excrete uric acid as encapsulated uric acid crystals through bird's cloaca. Encapsulated uric acid crystals appear as round smooth spheres of varying sizes as those identified in our study, surrounded by a protein material. In the case of pigs, this type of excretion does not exist, and so manure particles were fragmented, rough, and angular particles, instead of rounded, spherical, and smooth particles. Feddes *et al.* (1992) described crystals of uric acid from turkey housing, as round spheres from 3 to 8 μm in diameter, and other fecal particles as similar to feed particles with varying sizes from 3 to 7 μm in diameter.

The three types of feed particles dominant in the feed source samples were probably related to different feed components: mineral particles (geometric salt-like), and more grain-like organic particles (angular, cracked, fragmented particles) could be found. Outside particles were mainly constituted of salt-like crystals and crustal material. Crustal material (cracked, fragmented particles) were comparable to soil erosion and dust particles (Skogstad *et al.*, 1999) typical from agricultural environments where livestock houses are located. The rest of the described particle types (hair, skin, and wood shavings) were generally consistent with the known standards (McCrone, 1992) and coherent amongst livestock categories and PM fractions.

Similarities amongst particle types belonging to different sources were due to the presence of small, irregular, fragmented, angular particles. These types of particles, however, were mostly layered in manure (mainly from pigs), and cracked and fragmented in feed and outside source. These differences could be useful in their classification. The main differences amongst sources were found amongst hair and skin and the rest of sources, because these presented the most well defined and homogeneous particle types and morphologies. The presence, however, of very similar geometric quadrangular, bar-shaped or cubic particles in feed and outside source could complicate further their morphological discrimination.

Chemical (elemental) composition of the different sources obtained from 3303 particle EDX analysis showed presence of similar elements in all sources because most sources in livestock houses have an organic origin, but different relative elemental concentrations. Presence of N, Na, Mg, Al, Si, P, S, Cl, K, and Ca were identified in all sources. Most of these are common elements found in biological structures (feathers and skin). Other elements like Al and Si are common in mineral dusts (feed, and consequently in manure, and outside source). Therefore, a clear difference between minerals (rich in Al, Si, and Ca) and organic particles (rich in N, Na, S, Cl, and Ca) could be seen. This difference could be made between feed and outside particles (mineral) and the rest of sources (organic). Other studies have reported similar elements present in PM from livestock houses. Using SEM-EDX, Aarnink *et al.* (2004) in pigs and Cambra-López *et al.* (2008) in rabbits also reported presence of similar elements. In broilers and fattening pigs using bulk analysis, higher levels of N and P in fecal particles compared with

skin and feed, and higher levels of N, K, Cl in skin compared with fecal particles and feed were identified (Aarnink *et al.*, 1999). The strong presence of P in airborne particles from manure was also reported by Schneider *et al.* (2001). Feddes *et al.* (1992) indicated that particles from fecal origin had higher K than particles from feed. As regards mineral particles, high levels of Al and Si have also been reported in crustal material (Esbert *et al.*, 2001; Shi *et al.*, 2003). The presence of metallic trace elements could be explained by the use of some of these elements as feed supplements to improve health and feed efficiency (Bolan *et al.*, 2004). Metals can be added to diets to enhance weight increase, prevent disease, and increase egg production in poultry. They can be used in antibiotics, coccidiostats, Cu compounds as fungistat or growth promoters in pigs and poultry, and Fe and Zn added as mineral supplements (Bolan *et al.*, 2004; Kelley *et al.*, 1996). Traces of As, Cd, Co, Cr, Cu, Fe, Mn, Mo, Ni, Se, Pb, and Zn have been found in poultry bedding rice-hulls and in wood shavings (Kpomblekou-A *et al.*, 2002; López-Mosquera *et al.*, 2008), and consequently in the litter in poultry. Differences in elemental concentrations amongst sources could be used by the discriminant analysis to distinguish amongst them.

Particle size varied amongst sources, and mainly depended on its mineral or organic origin. Generally disintegration particles from feed and outside source showed smaller sizes, compared with biological structures (feathers, hair, skin, and wood shavings), which were mainly bigger than 4 μm in diameter. Using SEM, the D_p of the particles calculated from the particle area, resulted in D_p higher than 2.5 μm in fine PM. This high figure could be explained by two facts: the first related to the D_p being the diameter in the two-dimensional view, parallel to the plane of the filter; and the second related to the differences between geometric diameter and aerodynamic diameter. As most particles showed irregular shapes, particles would impact on the filter in their most stable orientation, generally exposing the biggest dimension on the filter plane, thus possibly explaining these high figures in D_p (Conner *et al.*, 2001). The geometric diameter of particles is related to its aerodynamic diameter through a dynamic shape factor, which varies with the resistance force of the particle to a fluid motion (Davies, 1979). Therefore, elongated particles (fibrous-like) which can show their longest axis in the direction of the flow, or large and thin (flake-like) particles with

low densities, could place small resistance to it, and they could be aerodynamically separated during sampling into a smaller diameter than they would if they were separated by their geometric diameter. Consequently, the accuracy of sizing particles using SEM can be reduced, as particles deviate from spheres (Willis *et al.*, 2002).

All sources showed the highest particle counts in the lowest size ranges. This differed when expressed in mass. The experimental dust generation process was successfully applied to re-create the processes leading to airborne PM occurring in the livestock houses, as our results on particle size were comparable with those from on-farm measurements in other studies. For instance, Heber *et al.* (1988) determined more than 50% of particles from pig houses were smaller than 2.7 µm, showing higher particle counts in the smallest size ranges for grain meal than for starch, where most particles were found to be greater than 5.4 µm. Our results suggest that most of the generated particles from our feed samples could come from grain meal rather than from starch. Furthermore, starch agglomerates, which present a specific and identifiable morphology in the SEM (viewed as polyhedral or sub-spherical agglomerate grains) according to McCrone (1992) were rarely seen in the analyzed particles from feed in our study. Measured number PSD in the air of livestock houses have been described elsewhere and have been also identified as bi-modal (Lammel *et al.*, 2004; Schneider *et al.*, 2006). Our results on size distributions could be furthermore useful to compare with on-farm measurements, to identify similarities and differences between on-farm PSD and those from known sources, taking into account differences in the measurement instruments used.

Overall, the complete characterization gave similar results in fine and coarse PM, showing similar morphological and chemical characteristics in both factions. During the dust generation process, an insight of the dust potential (Miller and Woodbury, 2003) of the different sources was achieved. The variable amount of sample and the dust generation time needed to maximize number of particles collected on the filter suggested feathers and manure were readily aerosolized, and thus showed higher dust potentials compared with the rest of sources. Our results suggest that dried manure and feathers could easily become airborne under on-farm conditions, when exposed to air movement. Thus, manure and feathers could potentially have a direct

effect on the generation, concentrations, and emissions of PM in and from livestock houses. However, this aspect should be confirmed with specific source-apportionment studies in livestock houses, or by comparison of on-farm samples to source morphologies and chemical compositions presented in this study. With the present particle source characterization, it will be possible to assign unknown particles from the air in livestock houses to a separate source.

As health effects may be related to PM characteristics other than mass, data on particle morphology and chemical composition presented in this study could be also useful to understand effects of PM in the respiratory system. These data could be further used to assess human and animal exposures to PM and its constituents in livestock houses.

3.5.   Conclusions

1.   Distinct particle morphologies were identified in collected sources from different housing systems for poultry and pigs. Detailed source profiles (morphological and chemical) for known sources were developed.

2.   Qualitative description of particle types revealed unique morphological features related to each source and different particle morphological types related to livestock production systems. Digital image analysis software could be useful to extract such charactristics and quantify further differences.

3.   Although presence of N, Na, Mg, Al, Si, P, S, Cl, K, and Ca were identified in all sources, their relative elemental concentrations varies amongst sources and can be used to discriminate amongst them.

4.   With the average elemental concentrations presented in this study, the relative concentrations of P, N, Cl, S, K, Si, Na, Al, Ca, Mg, Sn, and Pb are useful for discriminating amongst sources in fine PM. The relative concentrations of P, N, K, S, Cl, Al, Ca, Cr, Na, Mg, Ba, and Fe are useful for discriminating amongst sources in coarse PM.

5.   Particle size varies amongst sources (from 2.1 to 18.1 μm projected area diameter), and mainly depends on its mineral or organic origin. Generally disintegration particles from feed and outside show smaller

sizes, compared with biological structures (feathers, hair, skin, and wood shavings), which are mainly coarse.

6. The described source specific particle-size distributions can be useful to compare with on-farm measurements, to identify similarities and differences between on-farm PSD and those from known sources.

7. Comprehensive particle characterization and complete source analysis was achieved including different housing systems for poultry and pigs in size-fractioned PM. The data presented herein and the developed source profiles will be useful to assign airborne PM samples and individual particles to known sources and to improve source identification and quantification in livestock houses, a preliminary step to develop specific strategies for its reduction.

3.6.   Acknowledgements

3.7.   References

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Chapter 4

Selection of particle morpho-chemical characteristics to use in source apportionment of particulate matter from livestock houses

Selection of particle morpho-chemical characteristics to use in source apportionment of particulate matter from livestock houses

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Abstract. Intensive poultry and pig houses are important sources of particulate matter (PM). The knowledge on the contribution of individual sources to PM in different fractions is essential to improve PM reduction from livestock houses. We investigated which input data (particle chemical, morphological or combined characteristics) were best to distinguish amongst specific sources of airborne PM in livestock houses. We used a cross-validation procedure with classification rules based on decision trees and analyzed misclassification errors. The PM from two livestock species (poultry and pigs), and in two different fractions (fine and coarse) was studied. Results showed the selection of the best input data varies depending on the sources, which depend on livestock species. Using only particle chemical characteristics results in higher overall classification accuracies than using only morphological characteristics. Particle morphological characteristics can make additional value when sources show distinctive and well defined morphologies or differ in size. Using combined chemical and morphological results in the highest overall classification accuracies (average of 69% of particles correctly assigned to their source) and lowest misclassification errors. The approaches presented in this study are promising to determine the contribution of different sources to PM in livestock houses and give insight in under and overestimation errors in the source apportionment.

Keywords: Livestock, Image analysis, Morphology, SEM-EDX, Source apportionment.

4.1.   Introduction

Intensive poultry and pig houses are important sources of particulate matter (PM) emissions, contributing to about 50% (poultry), and 30% (pig) of total PM emissions from agriculture in Europe (EMEP-CORINAIR, 2007). To protect the environment and to ensure health and welfare of humans and animals in and around livestock houses, the concentrations and emissions of PM within such buildings must be controlled.

To this end, it is essential to identify and quantify the individual contribution of each potential source to PM. In addition, information on size, morphology and chemical composition of individual particles offers the potential to specifically identify and quantify sources of PM (Casuccio *et al.*, 2004). Based on morphological and chemical particle characteristics, particles can be placed into classes (Kim and Hopke, 1988, en Wienke *et al.*, 1995). Single-particle analysis with scanning electron microscopy (SEM) can provide chemical and morphological descriptive characteristics from hundreds of individual particles which can be further used to classify particles into distinct classes which resemble sources (Kim and Hopke 1988a). To do this, each source must have distinctive morphological and/or chemical features, which can be used to discriminate between them. When this is not the case or very specific sources need to be apportioned and distinguished, detailed morpho-chemical source profiles are necessary, as well as adequate methods which can select the best variables for discriminating.

To tackle PM reduction in and from livestock houses, the focus is mainly on particles from biological sources. Particulate matter from livestock houses can be very variable, but it generally consists up to 90% organic matter (Aarnink *et al.*, 1999; Seedorf and Hartung, 2001). It is mainly composed of primary particles of biological origin, directly emitted from animal husbandry, containing substances such as manure, feed, feathers, skin, particles from bedding and including micro-organisms (germs, fungi, viruses, bacteria, toxins and allergens) (Aarnink *et al.*, 1999; Donham *et al.*, 1986; Feddes *et al.*, 1992; Heber *et al.*, 1988; Qi *et al.*, 1992). The knowledge on the contribution of each of these individual sources to PM in different fractions (fine, PM2.5 and coarse, PM10-2.5) is essential to improve PM

reduction in this field. Because most particles have a similar elemental composition, rich in C, O, N, P, S, Na, Ca, Cl, Mg, and K (Aarnink *et al.*, 2004; Cambra-López and Torres, 2008; Schneider *et al.*, 2001), this complicates application of source apportionment in livestock husbandry. However, Cambra-López *et al.*, (2010) reported that, although similar elements could be present in all sources, their relative elemental concentrations vary amongst sources and this can be used to discriminate between them. Furthermore, individual particles from different sources can show unique morphological features. The use of an automated system to extract such features can be useful to identify similarities and differences amongst sources (Cambra-López *et al.*, 2010). To quantify the contribution of sources of PM in livestock houses, it is important to look for the most efficient and accurate way to distinguish between them.

Data obtained from particle analysis can be analyzed systematically by expert systems. Expert systems can be applied as knowledge-engineering tools in any field to interpret, predict, diagnose, design, plan, monitor, and control systems (Hopke, 1991; Kim and Hopke, 1988). Furthermore, expert systems can develop custom rules in the form of a decision tree, based on examples with known variables and classes; and then classify according to their rules. The rule-generator programs search the features for which they can best separate one class from the others.

The aim of this work was to investigate which input data (particle chemical, morphological or combined characteristics) were best to distinguish amongst specific sources of airborne PM in livestock houses. PM from two livestock species (poultry and pigs), and in two different fractions (fine and coarse) was studied. The convenience of using each input data was analyzed using a cross-validation procedure with classification rules based on decision trees. The overall accuracy of the classification, and the underestimation and overestimation errors were calculated for each source. Its implications for use in source apportionment studies are discussed. With this information, individual apportionment to specific sources in livestock houses will be improved.

4.2. Material and methods

Fine (PM2.5) and coarse (PM10-2.5) PM source samples from poultry and pig houses were used in the assessment. We tested three scenarios to select the best input data to distinguish between specific sources of airborne PM in poultry and pig houses: firstly classification using only particle chemical characteristics; secondly, classification using only particle morphological characteristics; and thirdly, the combination of both data sets.

Figure 4.1 shows examples of apportioning of particles to certain sources, chemically or morphologically. Examples: (a) one spherical agglomerate from manure (top) and one long-thin particle from feathers (bottom) in poultry showing different elemental composition and morphology; (b) two particles showing very similar elemental composition and morphology but belonging to different sources in pigs, manure (top) and feed (bottom); (c) two particles showing very similar morphologies but different elemental composition, one from feathers (top) and one from wood shavings (bottom); and (d) two particle showing very similar elemental compositions (rich in Na and Cl) but different morphology belonging to different sources in pig feed (top) and outside pig houses (bottom).

Single particle chemical and morphological characteristics were obtained using scanning electron microscopy (SEM) combined with energy-dispersive X-ray analysis (EDX). Single particle chemical and morphological data were obtained from particles from homogeneous known source samples. These data were used separately to develop a set of rules. The same particle data used to develop the set of rules were then used to test them following a cross-validation procedure. In this procedure, each particle (from a known reference source) was assigned to one of the sources applying the classification rules. The accuracy of the particle source assignment (correct particle classification) was evaluated through error matrices. A scheme showing the procedure used in this study is shown in Figure 4.2.
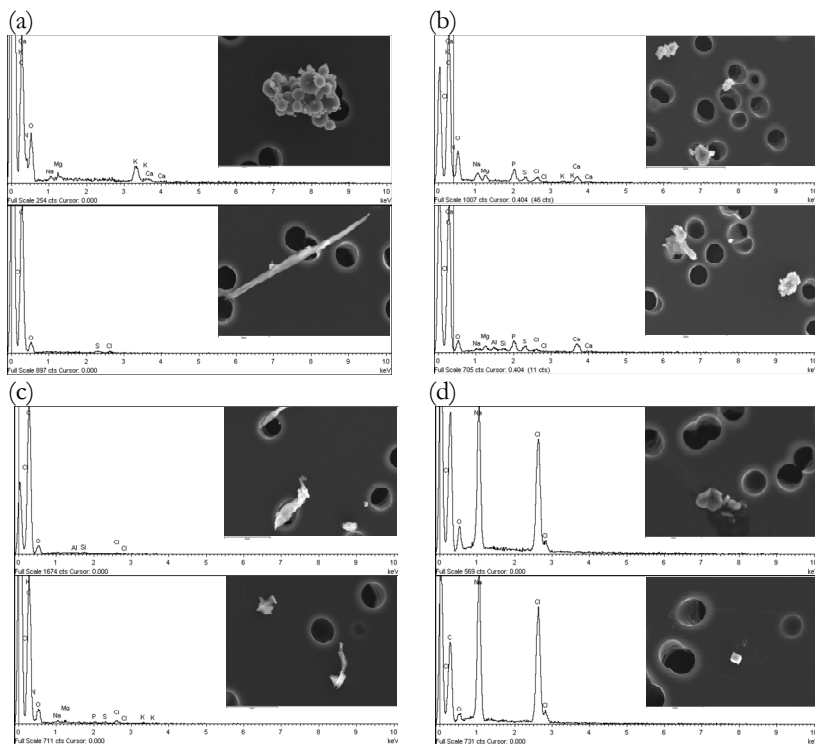
Figure 4.1. Examples of scanning electron microscopy photomicrographs of particles and elemental spectra showing chemical and morphological similarities and differences amongst sources of PM from poultry and swine houses. (a) Particle from poultry manure (top) and one long-thin particle from feathers (bottom); (b) particle from pig manure (top) and from feed manure (bottom); (c) particle from turkey feathers (top) and from wood shavings (bottom); and (d) particle from pig feed (top) and from outside source (bottom). Magnification 3000 to 3500x, scale bar 10 μm. Note 5 μm diameter filter pores, shown as round dark holes.

4.2.1. Input data: single-particle SEM-EDX analysis

A total of 96 fine PM samples and the same for coarse PM from 48 known source samples collected at 14 different livestock locations for poultry (including broilers, laying hens in floor and aviary system and turkey production) and pigs (including piglets, growing-finishing pigs, and dry-pregnant sow housings) were used in the assessment. Source samples were collected from major sources of PM (feathers, feed, hair, manure, skin, and wood shavings) at farm locations, obtained from previous study (Cambra-López *et al.*, 2010). These samples were dried, milled and aerosolized in a
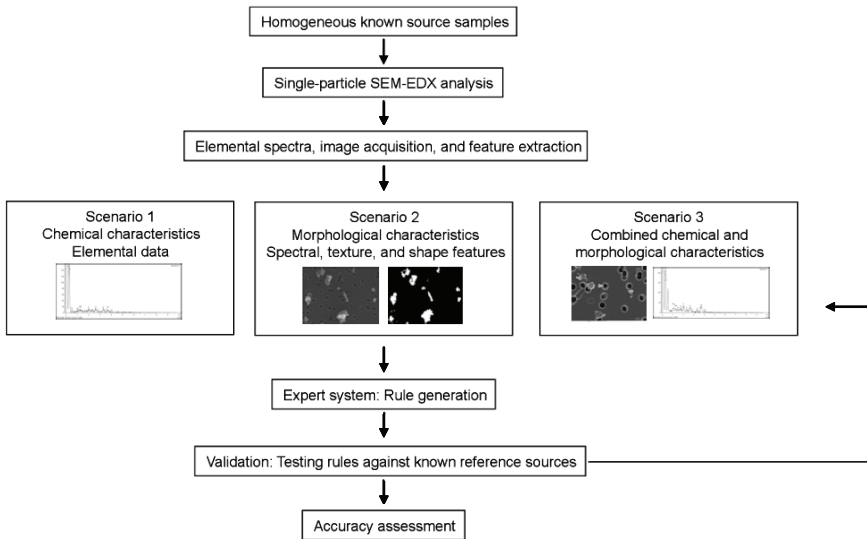
Figure 4.2. Flow diagram with the process used in the study.

dust generator. Besides, a representative sample of ambient outdoor fine and coarse PM at each location was collected on each sampling day. All PM samples were collected using a virtual cascade impactor (RespiCon, Wetzlar, Germany) on polycarbonate filters (37 mm Ø, 5 μm pore size). A portable pump (Genie VSS5, Buck Inc, U.S.) was used to suck air through the impactor at constant a flow of 3.11 L min^{-1}. Table 4.1 summarizes the origin of the data used in the assessment and the sources used in the analysis.

High-resolution SEM (JEOL, JSM-5410) combined with EDX (Link Tetra Oxford Analyzer) was used to obtain particle-by-particle chemical and morphological data. A small section (approximately 1 cm^2) of the as-collected polycarbonate filter from fine and coarse fractions was cut and mounted on a 12 mm carbon stub with a double-sided carbon adhesive tape. Samples were then coated with carbon using a vacuum evaporator to create a conductive coating for exposure to the SEM electron beam. At least three fields of view (spots) per filter sample were analyzed. On each analyzed field, both an image (photomicrograph at 1000x for coarse PM or 1800x for fine PM, saved in tif format 1024x768 resolution) and single particle X-ray spectra of every particle found in that field were obtained and stored. Within each field, the minimum projected area diameter for the

coarse particles was set at 1 µm. The minimum projected area diameter for the fine particles was set at 0.1 µm (Conner *et al.*, 2001). These limits were set because otherwise the detection and analysis of smaller particles was not reliable at the used magnifications. A total of 25 to 50 individual particles per sample were analyzed in each sample.

Table 4.1. Summary of sources used in the analysis for each livestock species.

Livestock species	Housing system	Source types
Poultry	Broilers - bedding	
	Turkeys - bedding	Feed Feathers
	Laying hens - floor	Manure Wood shavings
	Laying hens - aviary	Outside
Pigs	Piglets- slatted floor	Feed
	Growing-finishing pigs - partially slatted floor	Hair Manure
	Dry and pregnant sows - group housing	Skin Outside

4.2.1.1. Feature extraction

Particle chemical characteristics: Elemental data

Elements with atomic number ≥ 6 (carbon) were obtained from elemental x-ray spectra for each particle in each source. All spectra were confirmed and checked manually to correct for the contribution of the filter material (C and O). Based on chemistry, each particle was characterized by 25 elements (N, Na, Mg, Al, Si, P, S, Cl, K, Ca, Fe, Ni, Cu, Zn, Ag, Pb, Sn, Cr, Co, Ba, Br, Ti, V, Sb, Au). All elements were introduced in the expert system at once, because the decision tree approach can take into account correlation between variables, before applying rules.

Particle morphological characteristics: Spectral, texture, and shape features

The stored images (SEM photomicrographs of each field of view) were analyzed using the Object Based Image Analysis (OBIA) approach (Blaschke, 2010) using FETEX 2.0 Software (Ruiz *et al.*, 2010). All images were radiometrically corrected by background value adjustment to avoid spectral differences due to acquisition conditions, and to equalize the background value to compare intensity values between images. Individual

particles were defined by means of segmentation using thresholding. The OBIA software extracted both image and shape based features for each detected particle (object): spectral and texture features (image based), and morphological features (shape based).

Spectral features provided information about the spectral response of particles through their grey level (intensity) properties. Texture features provided information about the spatial distribution of the intensity values in the image, giving information about heterogeneity, contrast, and rugosity of particles. These features were uniquely referred to an object, extracted from the group of pixels that constituted a particle (Balaguer *et al.*, 2010). Histogram-based (kurtosis and skewness) features and seven of the most commonly used texture features based on the grey level co-occurrence matrix proposed by Haralick *et al.* (1973) were extracted. Finally, also as texture features, the mean and the standard deviation of the edgeness factor, representing the density of edges present in the neighborhood of each pixel (Laws, 1985) were extracted. Morphological features provided information about the complexity in the shape of the particles. Particle projected area, perimeter, and ellipse semi-axis values were extracted. Based on ratios between the area and the perimeter of the particles, compactness (C) (equation 1), shape index (SI) (equation 2), and fractal dimension (FD) (equation 3) were calculated. Based on morphological characteristics, each particle was characterized by 23 variables, summarized in Table 4.2.

$$C = \frac{4 \times \pi \times Area}{Perimeter^2} \tag{1}$$

$$SI = \frac{Perimeter}{4 \times \sqrt{Area}} \tag{2}$$

$$FD = 2 \times \frac{\log\left(\frac{Perimeter}{4}\right)}{\log(Area)} \tag{3}$$

where:
Perimeter is the length of the outline of a particle surrounding the area.
Area is the surface of the particle.

The most meaningful morphological descriptive features were selected before being introduced in the expert system to avoid redundancy and

obtain an efficient object description. Correlation analysis was used to group and interpret the redundancies in the information provided by the analyzed morphological variables using SAS Software (2001). Correlation between the complete set of variables was computed and analyzed. With this information, non-explanatory variables could be removed from the analysis.

Table 4.2. List and description of morphological particle characteristics based on spectral, texture and shape features.

Morphological feature	Basis and description	Variables
Spectral	Grey level intensity properties of particles	Mean, standard deviation, minimum, maximum, and range of intensity
Texture	Histogram-based characteristics	Skewness and kurtosis
	Based on the grey level co-occurrence matrix	Contrast, uniformity, entropy, variance, covariance or product moment, inverse difference moment, and correlation
	Density of edges present in the neighborhood of each pixel	Mean and the standard deviation of the edgeness factor
Shape	Particle length and size	Area, perimeter, and ellipse semi-axis (axis A and B)
	Ratios between the area and the perimeter of the particles	Compactness, shape index, and fractal dimension

4.2.2. Expert system: User-defined classification rules

We used a rule-generator expert system to create classification rules based on decision trees from the single-particle data from homogeneous known source samples. For each livestock species (poultry and pigs) and in each scenario, chemical, morphological or combined characteristics were introduced in the system to generate rules. Note that although data source was the same for all three scenarios, the number of particle observations was not equal amongst scenarios because inherent EDX spectra correction and acquisition procedures and the detection limit using OBIA. Hair source was only included in the assessment using morphological characteristics (scenario 2) because it showed very high carbon and oxygen peak in the SEM-EDX which was confused with the background filter composition.

4.2.2.1. Rule generation based on decision trees

Classification rules based on decision trees were generated for each group of sources in a given livestock species (see sources in Table 4.1). Classification rules were generated separately for the different input data in each scenario,

separately for poultry and pig sources, and separately for fine and coarse PM. Consequently the process of generating rules was repeated 12 times. The process of building a set of rules in the form of a decision tree worked by dividing data using mutually exclusive conditions until the newly generated subgroups were homogeneous, i.e. all the elements in a subgroup belonged to the same source or a stopping condition was fulfilled. Decision trees used a hierarchical structure to develop the set of rules for each particle belonging to a known reference source, using organized conditions such as greater than, less than, equal to, addition, and subtraction to search the variables and conditions for which it could best separate particles from one source from the others with the given input data. Decision trees were built using See 5 Software, using the C5.0 classification algorithm, which is the latest version of the algorithms ID3 and C4.5 developed by Quinlan (1993). The C5.0 algorithm manages several data types, such as continuous or discrete, thus it is the most widely used to deduce decision trees for classifying images (Zhang and Liu, 2004). To improve accuracy, the boosting multi-classifier method was used, where the final classification rule results from the weighed average of ten decision trees, where the next decision tree corrects from the errors of the previous one (Freund, 1995).

4.2.2.2. Validation of classification rules against known reference sources

We used the jackknifing procedure (a form of cross-validation statistical method) to assess the accuracy of the classification rules and validate them against reference source data in each scenario. This method involves re-sampling data, by repeatedly applying the generated rules to the same sampled set of data used to create them. The jackkniffing procedure works by leaving out a single observation at a time (one particle), generating rules for the rest of the particles, and then validating those rules against the left out particle observation. This was done for all observations. As a result from this validation, the accuracy of the classification and the degree of misclassification among sources was analyzed using error matrices or contingency tables (Aronoff, 1982; Congalton, 1991; Story and Congalton, 1986).

The error matrix was built by comparing the source assigned to each particle observation after the cross-validation process with its reference source; and

it presented the number of times a correct particle source assignment was made. These steps were essential to assess how well the classification rules fitted to the reference source data. Error matrices were also used to analyze the degree and direction of the most frequent misclassifications and to understand better and predict how the future classification of airborne on-farm samples would work when applying these classification rules to a mixture of unknown particles.

As an example, the construction of the error matrix in a given scenario, for a given number of particles (N observations) from two sources (source 1 and 2), worked by classifying each observation into one of the sources, corresponding to one of the four cells in the error matrix (Table 4.3). The classification rules would assign each particle observation into source 1 or 2 depending on its characteristics (input data), which vary depending on the scenario. In the example below, a, b, c, and d are the observed particle frequencies of source 1 and 2. They add up to the sample size (N). The sum of reference particles, the row total (n_x), equals the frequency (total number of particles) actually belonging to each source. The sum of all classified particles, the column total (m_x), equals the frequency (total number of particles) classified into each source after cross-validation process. On the one hand, 'a' equals the number of times a particle belonging to source 1 was correctly classified into source 1; 'b' equals the number of times a particle from source 1 was misclassified into source 2; analogously, 'c' equals the number of times a particle belonging to source 2 was misclassified into source 1; and finally 'd' equals the number of times a particle belonging to source 2 was correctly classified into source 2. In other words, the number of particles 'b' should have been assigned to source 1; and the number of particles 'c' should have been assigned to source 2. Cell 'b' and 'c' are related in the way that 'b' represents the underestimation of source 1, as the number of particles omitted from source 1 and incorrectly assigned to source 2. Cell 'c' represents the overestimation of source 1, as the number of particles from source 2 incorrectly assigned to source 1.

Table 4.3. Example of error matrix or contingency table for N observation and two sources.

Reference	Classified as		
	Source 1	Source 2	Row total (n_x)
Source 1	a	b	n_1
Source 2	c	d	n_2
Column total (m_x)	m_1	m_2	N=(a+b+c+d)

Overall measure of accuracy was obtained by dividing the total correct validations in each source (diagonal cells in Table 4.3) by the total number of classified particles (N) (equation 4). Misclassifications were calculated as measures of underestimate and overestimate error. The sum of the number of particles that have been incorrectly assigned to the reference source divided by the row total represented the underestimate error for each source the row represented (equation 5 and 6). The sum of the number of particles that have been incorrectly assigned to the classified source divided by the column total represented the overestimate error for each source the column represented (equation 7 and 8). To compare results and analyze under and over estimations, error matrices were standardized by the reference number of particles in each source (n_x). This means that after standardization n_2 equals n_1. The prediction accuracy of source apportionment was finally calculated dividing the column total (m_x) by the row total (n_x) for each source (equation 9 and 10).

$$Overall\ accuracy = \frac{(a+d)}{N} \tag{4}$$

$$Underestimate\ error\ source\ 1 = \frac{b}{n_1} \tag{5}$$

$$Underestimate\ error\ source\ 2 = \frac{c}{n_2} \tag{6}$$

$$Overestimate\ error\ source\ 1 = \frac{c}{m_1} \tag{7}$$

$$Overestimate\ error\ source\ 2 = \frac{b}{m_2} \tag{8}$$

$$Prediction\ accuracy\ source\ 1 = \frac{m_1}{n_1} \tag{9}$$

$$Prediction\ accuracy\ source\ 2 = \frac{m_2}{n_2} \qquad\qquad (10)$$

We also estimated error matrices and overall accuracies based on particle mass instead of particle numbers (frequency). We calculated the particle mass in each source, in each livestock species and PM fraction using the particle-by-particle masses in scenario 2 and 3. The overall accuracy was then obtained by dividing the mass from each correct validation in each source by the total mass of all classified particles. Misclassification errors (underestimate and overestimate) were also calculated in the same way as for particle numbers. The mass for each particle was calculated from the area and diameter provided by the SEM images, assuming a value for particle density. Density values of 1.2 g cm^{-3} (feathers), 2.6 g cm^{-3} (feed), 1.3 g cm^{-3} (hair), 1.5 g cm^{-3} (manure and wood shavings), 1.4 g cm^{-3} (skin), and 2.1 g cm^{-3} (outside) were used (McCrone, 1992). Calculations in numbers and in mass were performed because as particles from each source can have different sizes and consequently different masses (Cambra-López *et al.*, 2010), the effect of correct classifications and misclassifications could differ.

4.3. Results

4.3.1. Scenario 1: Particle classification based only on chemical composition

A set of rules based on decision trees were developed using only chemical composition data from 1113 particles in poultry for PM2.5, and 1133 for PM10-2.5; and from 522 particles in pigs for PM2.5, and 535 for PM10-2.5. Overall accuracies of the generated rules using particle chemical characteristics were higher in pigs compared with poultry. Overall accuracies varied from 58 to 62% in poultry and from 64 to 73% in pigs, for PM2.5 and PM10-2.5.

In poultry (Table 4.4), average misclassification errors ranged from 37 to 43%. Manure source showed the lowest misclassification errors, being underestimate errors (from 8 to 14%) slightly lower than overestimate errors (from 21 to 26%). Wood shavings source showed the highest misclassification errors, being underestimation errors higher than overestimate errors: approximately 63 to 65% of particles from wood

shavings were omitted from its reference source (underestimate error) and incorrectly assigned to other sources, but only 35 to 43% of particles from other sources were incorrectly assigned to wood shavings (overestimate error). The rest of sources presented similar underestimate and overestimate errors which varied from 33 to 63%, being the highest in outside source in PM10-2.5. Overall, results were similar for PM2.5 and PM10-2.5.

In pigs (Table 4.5), average misclassification errors ranged from 25 to 36%. All sources showed, compared with poultry, low misclassification errors (ranging from 10 to 53%) except for outside source in PM10-2.5 which presented a high underestimate error (72%). Feed and manure sources showed higher overestimate than underestimate errors; whereas skin and outside sources showed higher underestimate than overestimate errors for PM2.5 and PM10-2.5.

Table 4.4. Underestimate error (UE) and overestimate error (OE) per source and average, in percentage (%) number of particles, for poultry, for PM2.5 and PM10-2.5, using only particle chemical composition.

Reference source	PM2.5		PM10-2.5	
	UE	OE	UE	OE
Feathers	32.5	44.8	34.2	46.7
Feed	33.5	46.6	38.9	47.2
Manure	13.8	21.1	8.3	25.5
Wood shavings	62.9	34.7	64.5	43.3
Outside	47.5	39.7	63.0	50.6
Average	38.0	37.4	41.8	42.7

Table 4.5. Underestimate error (UE) and overestimate error (OE) per source and average, in percentage (%) number of particles, for pigs, for PM2.5 and PM10-2.5, using only particle chemical composition.

Reference source	PM2.5		PM10-2.5	
	UE	OE	UE	OE
Feed	25.1	45.4	28.5	53.1
Manure	9.9	17.0	10.8	21.3
Skin	31.0	14.8	33.3	19.8
Outside	44.2	23.9	72.0	44.9
Average	27.6	25.3	36.2	34.8

4.3.2. Scenario 2: Particle classification based only on morphological characteristics

Another set of rules were developed based on decision trees, using only morphological characteristics from 1400 particles in poultry for PM2.5, and 1617 for PM10-2.5; and from 599 particles in pigs for PM2.5, and 697 for PM10-2.5. Overall accuracies of the generated rules using particle

morphological characteristics were higher in pigs compared with poultry, and mostly lower than in scenario 1. Overall accuracies varied from 40 to 47% in poultry and from 60 to 67% in pigs, for PM2.5 and PM10-2.5.

In poultry (Table 4.6), average misclassification errors ranged from 53 to 60%. In number of particles, misclassification errors were generally higher than 40% in all sources in PM2.5 and PM10-2.5. Only manure source showed lower misclassifications, being underestimate errors (from 19 to 39%) lower than overestimate errors (from 55 to 56%). Wood shavings and outside source showed high underestimate errors in PM2.5 (above 70%). In particle mass, feed and outside sources showed higher underestimate and overestimate errors than in number of particles. Mass from feed and outside sources showed especially high underestimate errors (90 to 96%), but also high overestimate errors (94%) in feed in PM10-2.5.

In pigs (Table 4.7), average misclassification errors ranged from 30 to 42%. In number of particles in PM2.5 and PM10-2.5, misclassification errors were lower than in poultry. Hair source showed very low misclassifications expressed as low underestimate and overestimate errors (ranging from 3 to 18%). Manure source also showed low underestimate errors (from 8 to 14%) but presented higher overestimate errors (from 46 to 49%) compared with hair, consequently showing more particles from other sources incorrectly assigned to manure source. On the contrary, skin source showed higher underestimate errors (from 24 to 41%) than overestimate errors (7 to 14%). Overall, feed and outside sources showed the highest misclassification errors. In particle mass, feed and outside source showed generally higher misclassification errors than in number of particles. Underestimate errors of feed and outside were much higher (from 70 to 98%) compared with overestimate errors (from 12 to 70%). The mass of particles from hair showed few misclassifications. Skin source showed totally different results in mass compared with numbers, showing higher overestimate (41 to 54%) than underestimate errors (3 to 4%) in mass.

Table 4.6. Underestimate error (UE) and overestimate error (OE) per source and average, in percentage (%) per number and mass, for poultry, for PM2.5 and PM10-2.5, using only morphological characteristics.

| Reference source | PM2.5 | | | | PM10-2.5 | | | |
| | Number | | Mass | | Number | | Mass | |
	UE	OE	UE	OE	UE	OE	UE	OE
Feathers	40.5	64.5	20.7	57.4	49.0	61.2	16.9	44.2
Feed	61.5	60.6	89.6	40.9	70.2	60.7	95.8	94.3
Manure	39.3	54.9	19.5	63.5	19.1	56.2	15.0	51.6
Wood shavings	86.9	55.0	77.7	65.4	69.8	41.7	45.3	34.5
Outside	70.9	58.9	93.3	39.4	68.5	44.4	90.2	47.0
Average	59.8	58.8	60.2	53.3	55.3	52.8	52.6	54.3

Table 4.7. Underestimate error (UE) and overestimate error (OE) per source and average, in percentage (%) per number and mass, for pigs, for PM2.5 and PM10-2.5, using only morphological characteristics.

| Reference source | PM2.5 | | | | PM10-2.5 | | | |
| | Number | | Mass | | Number | | Mass | |
	UE	OE	UE	OE	UE	OE	UE	OE
Feed	50.0	48.4	92.8	69.4	39.3	46.2	69.8	11.7
Hair	2.9	5.3	0.0	30.0	16.7	5.7	20.3	5.4
Manure	14.3	45.5	9.2	42.1	7.8	48.7	10.9	46.9
Skin	40.7	7.1	4.2	40.9	23.8	13.7	2.7	53.9
Outside	56.2	45.0	89.6	29.1	86.9	57.2	98.3	33.9
Average	32.8	30.3	39.2	42.3	34.9	34.3	40.4	30.4

4.3.3. Scenario 3: Particle classification using combined data set (both chemical and morphological characteristics)

The last set of rules were developed based on decision trees, using both chemical composition and morphological data from 618 particles in poultry for PM2.5, and 805 for PM10-2.5; and from 317 particles in pigs for PM2.5, and 337 for PM10-2.5. Overall accuracies of the generated rules using both chemical and morphological characteristics were higher in pigs compared with poultry, and higher than in scenario 2. Overall accuracies varied from 58 to 68% in poultry and from 72 to 78% in pigs, for PM2.5 and PM10-2.5.

In poultry (Table 4.8), average misclassification errors ranged from 30 to 42%. In number of particles, most sources showed misclassification errors varying from 25 to 60% in PM2.5 and PM10-2.5, except for manure source. Manure source showed the lowest misclassifications, and presented higher overestimation errors (from 23 to 26%) than underestimate errors (from 6 to 15%). Wood shavings source showed the highest misclassification errors showing much higher underestimate errors (from 60 to 77%) than overestimate errors (from 18 to 44%). In particle mass, misclassification

errors for wood shavings source in PM10-2.5 were lower compared with number of particles. In particle mass, outside source presented very high underestimate error (96%) in PM10-2.5. For the rest of sources, misclassifications results were generally comparable in particle mass and in number.

In pigs (Table 4.9), average misclassification errors ranged from 21 to 30%. In number of particles, all sources except for outside source in PM10-2.5 showed low misclassifications expressed as low underestimate and overestimate errors (ranging from 7 to 45%) in PM2.5 and PM10-2.5. In particle mass, skin source showed much higher overestimate errors (from 23 to 31%) than underestimate errors (1%). Mass of skin followed the same trend as in scenario 2, presenting opposite results in number of particles compared with mass as regards over and underestimation. For the rest of sources, results were generally comparable in particle mass and in number.

Table 4.8. Underestimate error (UE) and overestimate error (OE) per source and average, in percentage (%) per number and mass, for poultry, for PM2.5 and PM10-2.5, using combined chemical and morphological characteristics.

| Reference source | PM2.5 | | | | PM10-2.5 | | | |
| | Number | | Mass | | Number | | Mass | |
	UE	OE	UE	OE	UE	OE	UE	OE
Feathers	29.1	53.2	18.0	58.7	24.8	44.5	11.2	32.1
Feed	49.4	39.3	49.2	13.1	27.1	34.7	43.4	60.1
Manure	15.3	26.0	10.9	23.5	5.9	23.4	5.3	22.3
Wood shavings	76.6	43.7	71.3	40.5	60.3	17.9	23.7	20.7
Outside	38.6	43.7	30.4	11.2	43.0	30.1	95.7	53.4
Average	41.8	41.2	36.0	29.4	32.2	30.1	35.9	37.7

Table 4.9. Underestimate error (UE) and overestimate error (OE) per source and average, in percentage (%) per number and mass, for pigs, for PM2.5 and PM10-2.5, , using combined chemical and morphological characteristics.

| Reference source | PM2.5 | | | | PM10-2.5 | | | |
| | Number | | Mass | | Number | | Mass | |
	UE	OE	UE	OE	UE	OE	UE	OE
Feed	25.0	35.4	45.3	32.6	10.8	45.3	65.8	49.2
Manure	8.9	19.0	11.1	34.1	7.0	18.5	6.4	13.4
Skin	21.1	6.5	0.5	22.9	22.6	6.9	0.5	30.7
Outside	33.3	23.3	52.6	13.7	70.2	24.7	39.0	24.8
Average	22.1	21.1	27.4	25.8	27.7	23.9	27.9	29.5

4.4.   Discussion

In general terms, overall accuracies were higher when using only particle chemical characteristics (scenario 1) compared with scenario 2 (morphological characteristics); whereas the highest accuracies were obtained using scenario 3 (combined chemical and morphological characteristics). This indicates that PM from livestock houses comprises a wide range of particle types not only between but also within sources, as it has been reported in other studies (Cambra-López *et al.*, 2010). This makes it difficult to find a single feature (based on chemical or morphological characteristics only) that can distinguish one source from the rest as a rule of thumb. For this reason, results in scenario 3 showed higher overall accuracies and lower misclassification errors compared with the other scenarios. In this scenario, the classification rules could search for the best criteria for classification from a wider range of options, using chemical characteristics when sources were more similar morphologically, and morphological characteristics when sources were more similar chemically. Therefore, the selection of the best input data can vary depending on the sources, which depend on livestock species. Our results suggest livestock species can be an important variation factor because each of the three scenarios performed differently for poultry compared with pigs. In our study, only feed, manure, and outside source were common in poultry and pig tests.

In poultry, higher accuracy and lower misclassifications were observed in scenario 1 compared with scenario 2, while in pigs scenario 1 and 2 performed more similarly. These results indicate that most sources in poultry houses are best differentiated by their chemical composition instead of by their morphological characteristics. This could be influenced by the strong presence of P and K in particles from manure in poultry compared with the rest of sources (Cambra-López *et al.*, 2010; Schneider *et al.*, 2001) resulting in rather stable and homogeneous elemental composition of manure from poultry, as regards its more diverse and complex morphology. The higher misclassification errors in scenario 2 compared with scenario 1 for the manure source in poultry, could be explained by the existence of two types of manure particles from poultry's excreta. Feddes *et al.* (1992) reported the presence of these two morphological types of particles in

poultry excreta: rounded spheres from 3 to 8 μm in diameter, and other less rounded and more irregular fecal particles in turkeys. Furthermore, particle size could also explain the high misclassification errors in scenario 2 in poultry compared with scenario 1. Cambra-López *et al.* (2010) reported a smaller range for particle size (expressed as projected area diameter) in particles from poultry sources than from pig sources. For instance, average particle's diameter of feathers, feed, manure, wood shavings, and outside was shown to vary between 2.1 and 5.9 μm; whereas particles from skin and hair (only present in pigs) can show diameters two-fold to three-fold bigger. The high underestimate error for wood shavings in scenario 2 (higher in PM2.5 compared with PM10-2.5) might be explained by the fact that particles from wood shavings in PM2.5 are smaller, and less elongated and fibrous than in PM10-2.5 and consequently could be easily confused with particles from other sources more. This could also be the reason why feed and outside sources generally presented higher misclassification errors in scenario 2 compared with scenario 1 (especially in poultry). These two sources have been reported to show irregular and angular morphologies and similar size and size distributions (Cambra-López *et al.*, 2010). Furthermore, our results show that size-only is not a recommendable variable to distinguish amongst most sources in livestock houses, because particles from different sources can be found in the same size ranges. Size can only be useful to distinguish amongst sources when one source with big particles (e.g. skin) wants to be distinguished from the rest. In pigs, the higher overall accuracy presented in scenario 2, compared with poultry, could have also been influenced by the very low misclassification errors for hair source, which can result from very distinctive and well defined individual particle morphology for this source.

The observed differences in misclassification errors between particle numbers and particle mass indicate two facts: (i) in sources showing small particles (e.g. feed and outside), big particles are more frequently misclassified into other sources than small particles; and (ii) in sources showing big particles (e.g. skin), small particles are more frequently misclassified into other sources than big particles. This could be seen in the higher underestimate errors in mass compared with numbers for sources showing generally small particles (feed and outside). Furthermore, our

results indicate that these misclassified particles (from feed and outside) were incorrectly assigned to sources showing big particles (such as skin), suggested by the higher overestimate errors in mass compared with numbers for skin source. Sources showing big particle masses (such as feathers and wood shavings in poultry, and especially hair and skin in pigs) presented higher overestimate than underestimate errors in mass compared with numbers suggesting it was probably small particles which had little influence on the mass which were misclassified. In mass, the effect of one single misclassification of a big particle could have more effect than a misclassification of a small particle, expressed in number. Nevertheless, to improve the understanding of misclassification and their influence in particle mass, the selection of particles should have been focused on coarse particles, and not on the whole size range as in this study.

The main objective of this study was to investigate which input data (particle chemical, morphological or combined characteristics) were more appropriate to distinguish amongst specific sources of airborne PM in livestock houses. This can help improve the knowledge on the most cost-effective input data to use. Our results suggest that this can depend on which source to apportion. When identification and quantification of the contribution of all individual sources to PM concentrations and emissions in livestock houses is the objective, a combination of chemical and morphological characteristics give high accuracies. However, obtaining complete particle characterization is time consuming and manual SEM-EDX single-particle analysis is laborious and expensive. Our results suggest that when only few sources want to be distinguished from the rest, the use of particle chemical or morphological particle characteristics as separate input data could give good results. However, this can only be applied in specific cases. For instance, if particles from manure want to be distinguished from the rest of sources, the use of only chemical particle characteristics would result in 86 to 92% of manure particles being correctly classified. If hair and skin want to be distinguished from the rest of sources as in pig houses, then the use of only morphological particle characteristics would result in 60% (PM2.5) to 76% (PM10-2.5) of skin particles, and 83% (PM10-2.5) to 97% (PM2.5) of hair particles being correctly classified. To distinguish feed from the rest of sources, which might be of interest when

evaluating the effect of certain reduction techniques which focus on "low-dust" feeding systems (Costa *et al.*, 2007; Dawson, 1990; Nannen *et al.*, 2005), according to our results, either using particle chemical characteristics or combined combination of particle chemical and morphological characteristics would result in 50 to 89% of particles from feed being correctly classified. To make a general recommendation for future studies, Table 4.10 presents a list of the sources analyzed in this study and the recommended scenario (the one that showed the lowest misclassification errors) according to our results. When misclassification errors differ between scenarios, recommendations are straightforward. However, when misclassification errors are similar amongst scenarios for a given source (for instance in outside source), more than one scenario can be recommended.

Nevertheless, based on our results, to apportion all individual sources to PM concentrations and emissions in livestock houses, we would recommend the use of combined chemical and morphological particle characteristics (scenario 3). In this scenario, an average overall accuracy of 69% (standard deviation of 6%) for particle number and mass in PM2.5 and PM10-2.5 was obtained. In other words, on average 69% of particles belonging to a mixture of sources were correctly assigned to their reference source based on their chemical and morphological characteristics. This accuracy can be considered reasonably good and it implies that only about 30% of the particles would be misclassified and incorrectly apportioned. The implications for source apportionment in livestock houses of this misclassification value are low, because the main aim of source apportionment in livestock houses is to provide knowledge on most important sources which can be used to develop new PM reduction techniques and optimize the existing ones. Therefore, this level of accuracy would be sufficiently high and would allow obtaining the overall picture of the major or dominant sources of PM in livestock houses.

Table 4.10. Check list of recommended scenario for particle identification from different sources.

Source	Scenario 1 Particle chemical characteristics	Scenario 2 Particle morphological characteristics	Scenario 3 Combined chemical and morphological particle characteristics
Feathers			X
Feed	X		X
Hair		X	
Manure	X	X (only in pigs)	X
Skin	X	X (only PM10-2.5)	X
Wood shavings	X		
Outside	X		X

Error matrices in this study were used to analyze the degree and direction of the most frequent misclassifications. Our results indicate that when applying classification rules to airborne on-farm samples, certain sources could be systematically under or overestimated. Table 4.11 and Table 4.12 summarize the estimated under or overestimation for each source in poultry and pigs for the recommended scenario 3, derived from Table 4.8 and Table 4.9. Although errors are inherent to all calculations, the results presented in this study can be used in such a way that under and overestimation errors can be better understood and corrected using these figures, taking into account, that in real conditions, the final under or over estimation will depend on the contribution of each source to the airborne PM sample.

Table 4.11. Prediction accuracy of source apportionment for poultry based on underestimate and overestimate errors when using scenario 3.

Reference source	PM2.5		PM10-2.5	
	Number	Mass	Number	Mass
Feathers	1.5	2.0	1.4	1.3
Feed	0.8	0.6	1.1	1.4
Manure	1.1	1.2	1.2	1.2
Wood shavings	0.4	0.5	0.5	1.0
Outside	1.1	0.8	0.8	0.1

Table 4.12. Prediction accuracy of source apportionment for pigs based on underestimate and overestimate errors when using scenario 3.

Reference source	PM2.5		PM10-2.5	
	Number	Mass	Number	Mass
Feed	1.2	0.8	1.6	0.7
Manure	1.1	1.4	1.1	1.1
Skin	0.8	1.3	0.8	1.4
Outside	0.9	0.6	0.4	0.8

4.5. Conclusions

From our work using feathers, feed, manure, wood shavings, and outside PM sources in poultry, and hair, feed, manure, skin, and outside PM sources in pigs, we can conclude that:

- The selection of the most appropriate particle characteristics (chemical, morphological or combined morpho-chemical characteristics) to distinguish amongst particles from different sources in livestock houses depends on the sources, which depend on livestock species.

- Using only particle chemical characteristics results in overall classification accuracies varying from 58 to 62% in poultry and from 64 to 73% in pigs; it can be useful to apportion specific sources such as manure from the rest. In this case, the use of only chemical particle characteristics would result in 86 to 92% of manure particles being correctly classified.

- Using only particle morphological characteristics results in overall accuracies varying from 40 to 47% in poultry and from 60 to 67% in pigs; it can make additional value to using only chemical characteristics when sources show distinctive and well defined individual particle morphology or differ in size.

- Using combined chemical and morphological particle characteristics results in overall accuracies varying from 58 to 68% in poultry and from 72 to 78% in pigs (average 69%); it is the recommended approach to apportion all individual sources to PM concentrations and emissions in livestock houses.

- Our results show that the different approaches used in this study are promising to determine the contribution of different sources to PM in livestock houses. Results in this study also give insight in under and overestimation errors in the source apportionment.

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Chapter 5

Particulate matter emitted from livestock houses: On-farm source identification and quantification

Particulate matter emitted from livestock houses: On-farm source identification and quantification

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Abstract. To identify and quantify the contribution of different sources to fine and coarse PM emissions from poultry and pig houses, we compared the chemical and morphological characteristics of fine and coarse PM from known sources collected from livestock houses with the characteristics of on-farm fine and coarse airborne PM. We used two methods to estimate source contributions: classification rules based on decision trees and multiple linear regression. Source contributions were calculated in particle numbers and then estimated in particle mass. Based on particle numbers, results showed that in poultry houses, most on-farm airborne PM originates from feathers (ranging from 4 to 43% in fine and from 6 to 35% in coarse PM) and manure (ranging from 9 to 85% in fine and from 30 to 94% in coarse PM). In pigs, most on-farm airborne PM originates from manure (ranging from 70 to 98% in fine and from 41 to 94% in coarse PM). The contribution of manure to on-farm airborne PM was higher in coarse PM in poultry, but higher in fine PM in pigs. Feed had a negligible contribution to on-farm airborne PM compared with the rest of the sources. Based on particle mass, big particles such as wood shavings and skin gain relative importance compared with numbers. In poultry, most on-farm airborne PM still originates from feathers (ranging from 15 to 63% in fine and from 3 to 46% in coarse PM) and manure (ranging from 7 to 81% in fine and from 36 to 97% in coarse PM), but in pigs, skin contributed to the highest mass (ranging from 13 to 91% in fine and from 39 to 86% in coarse PM). Results presented in this study improve the understanding of where PM comes from in different livestock housing systems. This can be valuable to choose the optimal PM reduction methods.

Keywords: Animal housing, Dust, Emissions, Source apportionment.

5.1.   Introduction

Large amounts of particulate matter (PM) are emitted from livestock houses, which can compromise animals' and humans' respiratory health (Donham, 2000; Radon *et al.*, 2001; Zuskin *et al.*, 1995) and the environment, as well. The scientific community and stakeholders (farmers and local authorities) are seeking technically feasible and economically viable solutions to reduce these emissions to comply with air quality regulations. Preventing dust release from its source not only reduces emissions from the animal house, but improves indoor climate, as well. To develop such reduction techniques, it is necessary to accurately identify and quantify sources which contribute to PM in livestock houses. A complete assessment can be achieved knowing number and mass contributions. An accurate knowledge on the relationship between particle mass and number contributions could be useful to understand health risks, and predict the effect of reduction techniques.

Analytical methods used to characterize PM such as microscopic analysis, can supply useful but limited data on particle or source chemical composition and morphological characteristics. To further identify and quantify source contributions, source apportionment models are used to determine emission sources and their contribution to ambient PM concentrations at specific monitoring sites, called receptors. These models are very versatile and they can apportion PM to sources by relating chemical and physical properties of the source, to the properties measured at the receptor site (Watson *et al.*, 2002).

Source apportionment models based on multivariate liner regression can be used to investigate this relationship (to relate chemical and physical properties of the source, to the properties measured at the site) and they permit quantitative source apportionment. Linear regression is used to estimate the relative contribution of each known source as the linear sum of products of source compositions and source contributions, based on predetermined specific, accurate, and detailed source profiles (Hopke, 1991). Furthermore, expert systems based on supervised methods can be used to analyze data systematically. Expert systems can be applied as knowledge-engineering tools in any field to interpret, predict, diagnose, design, plan,

monitor, and control systems (Kim and Hopke, 1988). Expert systems can be used to develop custom rules in the form of a decision tree based on examples or training samples with known variables; and then classify according to their rules. User-defined rules based on decision trees have been used to sort and classify particles based on large datasets (Hopke, 2008; Hopke and Song, 1997; Kim and Hopke, 1988; Wienke *et al.*, 1995). Based on known source profiles, rules could also sort and classify particles into predetermined and selected classes or sources.

The formation of PM in livestock houses, its concentrations, and emissions depend on many physical and biological factors such as kind of housing and feeding, animal type, and environmental factors (Takai *et al.*, 1998). Generated PM in livestock houses mainly originates from feed, manure, bedding, and animal's skin, and feathers (Aarnink *et al.*, 1999; Donham *et al.*, 1986; Feddes *et al.*, 1992; Heber *et al.*, 1988; Qi *et al.*, 1992). Attempts to identify and quantify sources of PM in livestock houses have been made although only limited data from specific production systems related to single livestock categories are available (Aarnink *et al.*, 1999; Aarnink *et al.*, 2004; Feddes *et al.*, 1992; Heber *et al.*, 1988; Honey and McQuitty, 1979; Qi *et al.*, 1992). Comparable source contributions between and within livestock categories for different sized-particles are needed. To this end, specific methodologies which include statistical methods to calculate source contributions, standardized measuring protocols, and comprehensive field studies to characterize PM morphology and composition in different size fractions need to be developed.

It is generally accepted that to apply source apportionment models in livestock houses, it is necessary to obtain particle chemical characteristics. However, the presence of similar chemical elements (C, O, N, P, S, Na, Ca, Cl, Mg, and K) in most of the sources related to livestock PM can complicate discrimination amongst them. Hence, the use of specific and detailed source profiles is necessary and is encouraged (Cambra-López *et al.*, 2010c). Cambra-López *et al.* (2010a) reported that besides chemical data, morphological particle characteristics could be useful in source apportionment in livestock houses, because in some cases, livestock-related PM can be more heterogeneous in size and morphology than in chemical composition. Furthermore, using combined chemical and morphological

particle characteristics generally achieves the most accurate results and can decrease misclassification errors amongst sources (Cambra-López *et al.*, 2010a). Therefore, using only chemical or combined chemical and morphological particle characteristics can be used to apportion single sources to on-farm airborne PM and improve the knowledge on the quantitative importance of the different PM sources in terms of number and mass contributions.

The objective of this study was to identify and quantify the contribution of different sources to fine (PM2.5) and coarse (PM10-2.5) PM emissions from livestock houses based on chemical and morphological characteristics of particles. A comprehensive list of livestock categories and housing systems was surveyed, including seven different housing systems: broilers in bedding system, laying hens in floor system, laying hens in aviary system, turkeys in bedding system, and piglets, growing-finishing pigs, and dry and pregnant sows in slatted floor system. The contribution from each source to PM was estimated in number and in mass by comparing the chemical and morphological characteristics of fine and coarse PM from each source, with the characteristics of fine and coarse airborne PM from the livestock houses. Two methods were used to estimate source contributions: classification rules based on decision trees and multiple linear regression. This study will provide a better understanding of PM origin, essential to understand better potential health and environmental hazards of PM, and to improve actual reduction programs applicable to livestock houses.

5.2.   Material and methods

To identify and quantify the contribution of different sources to fine and coarse PM emissions from seven different housing systems for poultry and pigs, we sampled airborne fine and coarse PM on-farms and collected samples from known PM sources. Two different locations were sampled for each livestock housing system in The Netherlands.

5.2.1.   Housing and animals

Table 5.1 describes surveyed livestock species, type of housing system, ventilation system, number of animals, and animal age, where airborne and

source samples were collected. All surveyed livestock houses used automatically distributed feeding systems with crumbles or pelleted feed.

Table 5.1. Description of surveyed livestock houses.

Livestock species	Housing system	Farm location	Ventilation	Number of animals	Age (weeks)
Poultry	Broilers - bedding	1	Tunnel	50,400	4
		2	Roof	2675	3
	Laying hens - floor	1	Tunnel	3850	71
		2	Tunnel	16,500	22
	Laying hens - aviary	1	Tunnel	24,712	71
		2	Tunnel	35,000	50
	Turkeys - bedding	1	Ridge	5,000	12
		2	Ridge	4,040	10
Pigs	Piglets- slatted floor	1	Roof	125	8
		2	Roof	75	9
	Growing-finishing pigs - partially slatted floor	1	Roof	120	16
		2	Roof	60	20
	Dry and pregnant sows - group housing	1	Roof	39	Diverse
		2	Roof	46	Diverse

5.2.2. On-farm airborne and source samples

Duplicate virtual cascade impactors (RespiCon, Wetzlar, Germany) were used in each farm to sample simultaneously airborne fine and coarse PM onto separate polycarbonate filters (37 mm Ø, 5 μm pore size). Portable pumps (Genie VSS5, Buck Inc, U.S.) were used to suck air through each impactor at a constant flow of 3.11 L min^{-1}. Sampling was conducted during morning (from 09:00 to 12:00) at each livestock house. Samples were taken near the exhaust in each farm. Sampling time varied from 5 to 60 minutes, adjusted to obtain particle loads of 5-20 μg particles cm^{-2} filter, to minimize particle overlap (Willis *et al.*, 2002). Background (outside) samples were taken upwind of livestock houses in the same way as indoor samples in all farms. Sampling time outside varied from 30 to 60 minutes.

Additionally, a light scattering system (DustTrak TM Aerosol Monitor, model 8520, TSI Incorporated, Shoreview, U.S.) was used for on-line continuous airborne PM10 concentration measurement inside and outside livestock houses. Sampling time was 30 to 60 minutes. One-minute values were recorded and stored. Temperature and relative humidity were also recorded during each sampling, both inside and outside the livestock house, using temperature and relative humidity sensors (Escort ilog data logger, Askey Leiderdorp, The Netherlands).

On each farm, potential PM sources were collected. Sampled sources were: concentrate feed (all farms), manure (fresh excreta in poultry and fresh feces in pigs), feathers (in poultry), and wood shavings used as bedding material (present only in broilers and turkeys). We also collected skin samples in pig houses, but only from sows because it was impractical to collect such source from younger animals (piglets and growing-finishing pigs). Approximately 200 to 500 grams of a representative sample of feed, manure, and wood shavings were collected, except for feathers, and skin, where 10 to 50 grams were collected in clean polyethylene bags. Each sample was dried for 12 h at 70ºC. Dried samples were crushed in a ball mill during 1.5 minutes at 250 rpm. Dried and milled samples were stored at room temperature and then airborne PM was generated in a laboratory dust generator to collect airborne fine and coarse PM samples from each source. The dust generator consisted of a stainless steel cylinder of 20 cm diameter and 30 cm high with an airtight lid, which had a mechanical agitation system and rotatory blades at the end. A varying quantity, from 0.2 grams (feathers) to 40 grams (feed) of milled source was used in the dust generator, and agitated at 200 rpm. The generated PM was collected using a virtual cascade impactor (RespiCon, Wetzlar, Germany) and portable pump (Genie VSS5, Buck Inc, U.S.) using polycarbonate filters (37 mm Ø, 5 μm pore size). Sampling time varied from 1 minute to 7 hours, depending on the amount of particles generated, aiming at particle loads of 5 to 20 μg particles cm^{-2} filter (Willis *et al.*, 2002). Filter samples were stored in sealed filter cassettes at room temperature (20-25ºC) before analysis.

5.2.3. Morpho-chemical analysis of airborne and source samples

High-resolution Scanning Electron Microscopy (SEM) (JEOL, JSM-5410) combined with energy-dispersive X-ray analysis (EDX) (Link Tetra Oxford Analyzer) was used to obtain particle-by-particle chemical and morphological data. A small section (approximately 1 cm²) of the as-collected polycarbonate filter from fine and coarse fractions was cut and mounted on a 12 mm carbon stub with a double-sided carbon adhesive tape. Samples were then coated with carbon using a vacuum evaporator, to provide electrical conductivity and create a conductive coating for exposure

to the SEM electron beam. Detection of elements with atomic number ≥ 6 (carbon) was obtained from elemental x-ray spectra.

The SEM-EDX was conducted manually, operated under the same conditions throughout the study: accelerating voltage 10 keV, working distance 15 mm, electron probe current of 3 nA, magnifications 1000x for coarse PM, and 1800x for fine PM, and X-ray acquisition time 60 s per particle. Secondary electron mode was used for particle location, measurement, analysis, and image acquisition.

At least three fields of view (spots) per filter sample were analyzed. On each analyzed field, both an image (photomicrograph at 1000x or 1800x, saved in tif format 1024x768 resolution) and single particle X-ray spectra of every particle found in that field were obtained and stored. Within each field, the minimum projected area diameter for the coarse particles was set at 1 µm. The minimum projected area diameter for the fine particles was set at 0.1 µm (Conner *et al.*, 2001). These limits were set because otherwise, the detection and analysis of smaller particles was not reliable at the used magnifications. For each airborne sample, a total of 50 to 75 particles were chemically analyzed in each duplicate sample. For each source sample, a total of 25 to 50 particles were chemically analyzed. All spectra were confirmed and checked manually to correct for the contribution of the filter material (C and O).

The stored images (SEM photomicrographs of each field of view) were analyzed using the Object Based Image Analysis (OBIA) approach (Blaschke, 2010) using FETEX 2.0 Software (Ruiz *et al.*, 2010). All images were radiometrically corrected by background values to avoid spectral differences due to acquisition conditions and to equalize the background value to compare intensity values between images. Individual particles were defined by means of segmentation using thresholding. The OBIA software extracted both image and shape based features for each detected particle (object): spectral and texture features (image based), and morphological features (shape based). Based on chemistry, spectral, texture, and morphological features, each particle was exhaustively characterized by 48 variables (Cambra-López *et al.*, 2010a).

5.2.4. Source apportionment methods

Fine and coarse source samples, as well as on-farm airborne fine and coarse PM samples from each livestock house were used in source apportionment using classification rules based on decision trees and multiple regression techniques. Single particle chemical and morphological characteristics obtained using SEM-EDX were used as data sources. Apportionment results were calculated in number, and then estimated in terms of mass. Results provided by the two methods were compared and discussed.

5.2.4.1. Classification rules based on decision trees

Decision trees were used to develop a set of rules for each group of sources from each livestock house. Both single particle chemical and morphological characteristics from known sources obtained using SEM-EDX were joined in a combined database and used in this process. Decision trees were built using See 5 Software, using the C5.0 classification algorithm, which is the latest version of the algorithms ID3 and C4.5 developed by Quinlan (1993). Decision trees were created following the boosting multi-classifier method (Freund, 1995). The rule-generator program searched the features that best separated one source from the other by dividing data using mutually exclusive conditions until the newly generated subgroups were homogeneous, i.e. all the elements in a subgroup belonged to the same class or a stopping condition was fulfilled. The developed rules using the known sources were then applied to classify airborne on-farm samples into one of the known sources, based on their chemical and morphological characteristics.

Accuracy of this method was tested through cross-validation, applying the rules to the same source samples and comparing the source assigned to each particle using rules with its reference source per farm. Overall measure of prediction accuracy for number of particles was obtained by dividing the total correct validations in each source by the total number of classified particles.

5.2.4.2. Multiple linear regression

Multiple linear regression analysis was also used to apportion airborne PM sampled on the farms to the known sources. Single particle chemical

characteristics from known sources obtained using SEM-EDX were used in this process. The average PM concentration of elements in fine and coarse airborne on-farm samples were used as dependent variables and the average fine and coarse PM concentrations of elements in each source were used as independent variables. All elements were included at once in the model using Genstat (Genstat Committee, 2008), following equation 1:

$$Y_{im} = \sum_{k=1}^{n} (f_{ikm} \times F_{ikm}) \qquad (1)$$

where:

Y_{im}= relative concentration of the ith element in collected airborne fine or coarse PM in the mth farm (average of duplicate samples)

f_{ikm}= number contribution of the ith element of the kth source to airborne fine or coarse PM in the mth farm. The sum of the fractions was set to 1.

F_{ikm}= average relative concentration of the ith element in the kth source in the mth farm

5.2.4.3. Mass estimation

Results from classification rules based on decision trees and multiple linear regression were given in particle numbers. Particle number contributions were transformed into mass contributions based on the average mass of particles in each source. The mass for each single particle (m) was calculated from the projected area diameter (D_p) provided by the SEM images, based on a density value and shape factor, following the equation for the mass of a particle (equation 2) (Ott *et al.*, 2008). From single-particle masses, average particle mass per source was calculated.

$$m = \rho_p \times v_p = \rho_p \times \left[\frac{4}{3} \times \pi \times r^3 \right] = \frac{\rho_p \times \pi \times \left(\frac{D_p}{S_v} \right)^3}{6} \qquad (2)$$

where:

m= particle mass

ρ_p= particle density

v_p= particle volume

r= equivalent radius of a spherical particle

D_p= projected area diameter, $D_p = 2 \times \sqrt{Area/\pi}$

S_v= Volume shape factor. Correction factor to convert (D_p) to equivalent volume diameter, defined as the diameter of a sphere having the same volume as the irregular particle.

We assumed all particles were spheroids in this calculation, whose volume could be estimated from the volume of a sphere. The volume shape factor (S_v) equals 1 for spheres (Noll *et al.*, 1988). Average values for density were 1.2 g cm^{-3} (feathers), 2.6 g cm^{-3} (feed), 1.3 g cm^{-3} (hair), 1.5 g cm^{-3} (manure and wood shavings), 1.4 g cm^{-3} (skin), and 2.1 g cm^{-3} (outside) (McCrone, 1992).

5.3. Results

5.3.1. On farm PM airborne measurements

Average PM10 concentrations measured using light scattering system, relative humidity and temperature measured inside and outside livestock houses are presented in Table 5.2. Values in the table represent sampling time averages over 5 to 60 minutes, and standard error between the two surveyed houses for the same livestock species.

5.3.2. Source identification

Sources were identified through individual particle morphologies based on SEM observations. Different types of particles collected from different livestock housing systems were identified by comparison to known standards (Cambra-López *et al.*, 2010c; McCrone, 1992). Figure 5.1 shows examples of particle types from different livestock housing systems. In broiler houses, a mixture of particles showing "fluffy" appearance probably from feathers and flattened agglomerates are shown in Figure 5.1a. Also bent, sharp-edged particles from wood shavings and spherical particles from

Table 5.2. Summary of average (Avg) PM10 measurements, temperature (T) and relative humidity (RH) inside (in) and outside (out) surveyed livestock houses. Standard error (SE) represents variation between both surveyed livestock houses for the same housing system.

Livestock species	PM10 in (mg m^{-3})		PM10 out (mg m^{-3})	
	Avg	SE	Avg	SE
Broilers	1.96	0.55	0.08	0.05
Laying hens- floor	3.94	0.69	0.03	0.00
Laying hens- aviary	3.06	1.54	0.03	0.00
Turkeys	2.32	0.99	0.08	0.05
Piglets	1.44	0.11	0.03	0.01
Growing-finishing pigs	1.27	0.35	0.03	0.01
Dry and pregnant sows	0.39	0.01	0.03	0.01

Livestock species	T in (°C)		RH in (%)		T out (°C)		R out (%)	
	Avg	SE	Avg	SE	Avg	SE	Avg	SE
Broilers	23.2	N.D.	81.6	N.D.	13.2	N.D.	50.6	N.D.
Laying hens- floor	16.2	1.7	74.8	0.5	10.3	0.4	74.4	18.8
Laying hens- aviary	15.6	3.2	70.4	3.2	10.3	0.4	74.4	18.8
Turkeys	19.4	2.5	63.3	7.0	11.3	0.1	54.3	0.2
Piglets	25.2	0.1	75.8	0.3	11.4	1.8	55.0	20.5
Growing-finishing pigs	21.9	0.8	62.3	9.0	11.4	1.8	55.0	20.5
Dry and pregnant sows	23.9	N.D.	75.6	N.D.	13.3	N.D.	34.5	N.D.

N.D. No data due to equipment failure in one of the farms.

excreta could be identified (Figure 5.1a). In laying hens, spherical particles from excreta were dominant in collected PM (Figure 5.1b) and also in aviary system (Figure 5.1c). In turkey houses, bent, sharp-edged particles and spherical particles from excreta were identified (Figure 5.1d). In piglet houses, deposited round grey, smoothed particles, as if melted were identified together with some bright layered manure particles (Figure 5.1e). A mixture of layered, grain-like manure particles and big flattened skin particles were collected from piglet houses (Figure 5.1f) and growing-finishing pigs (Figure 5.1g). Scarce and small particles and flattened, folded and big skin particle were collected from dry and pregnant sow houses (Figure 5.1h).
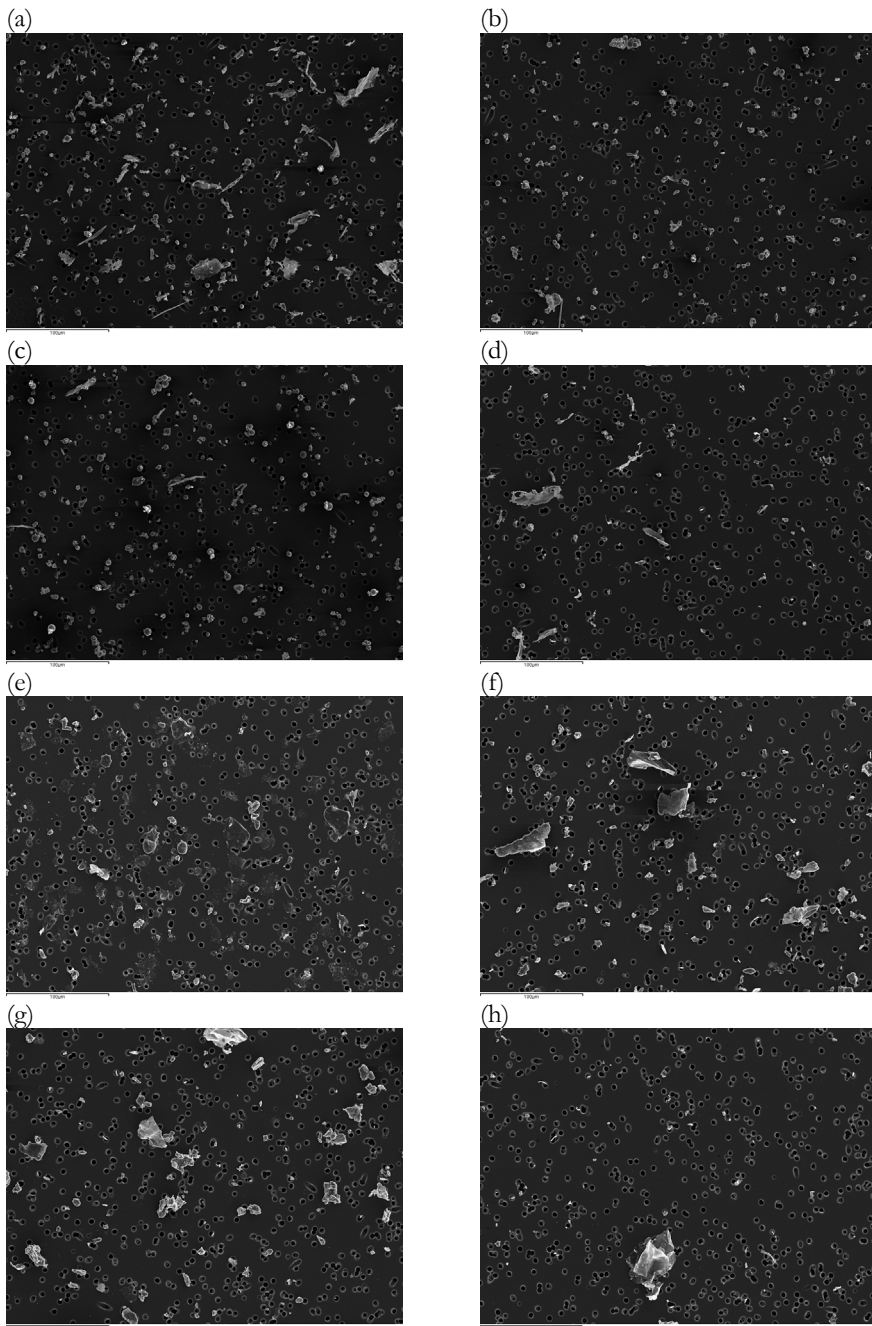
Figure 5.1. Examples of SEM images from on-farm airborne PM samples collected on polycarbonate filters (note 5 μm diameter filter pores shown as round dark holes). (a) Particles from broiler houses. Spherical particles from (b) laying hens- floor housing system and from (c) laying hens- floor housing

system. (d) Particles from turkey houses. (e and f) Particles from piglet houses. (g) Mixture of irregular shaped from growing-finishing pig houses. (h) Big skin particle collected from dry and pregnant sow houses. Scale bar 100 µm.

5.3.3. Source quantification

Source apportionment using classification rules based on decision trees and multiple linear regression resulted in percentage contributions of sources to on-farm airborne PM, expressed in particle numbers. A total of 912 individual particles were apportioned in fine and 1071 in coarse PM using classification rules based on decision trees. A total of 1546 individual particles were apportioned in fine and 1670 in coarse PM using multiple linear regression.

5.3.3.1. Contribution of sources to on-farm airborne PM expressed in number

Using classification rules based on decision trees

Results using classification rules based on decision trees are shown in Table 5.3 (fine PM) and Table 5.4 (coarse PM), together with method accuracies. Results indicated that in poultry, most of the PM originated from feathers and manure. Contribution of manure was generally higher in coarse PM (ranging from 30 to 87%) compared with fine PM (ranging from 9 to 85%). Manure contribution was higher in layer houses compared with broilers and turkeys; whereas feather contribution was higher in broilers and turkeys compared with laying hens. Where present, wood shavings contributed less than 20% of particle numbers. In pigs, most of the PM originated from manure. The contribution of manure was higher in fine PM (ranging from 70 to 89%) compared with coarse PM (ranging from 41 to 71%), for all pig categories. Skin and feed were the other most important contributing sources in pigs. Contribution of skin varied from 2 to 33%, varying between pig categories, being highest in coarse PM compared with fine PM in piglets and growing-finishing pigs, whereas being higher in fine PM compared with coarse PM in sows. Outside particles had a relevant contribution in broilers and turkeys, especially in fine PM; but also in coarse PM in sows. Contribution of feed was found below 16% for all livestock categories, being the highest in piglets, in both fine and coarse PM.

Standard errors of the estimated contributions between the surveyed livestock houses were generally low, except for some cases where a big difference in the contribution of the same source in different livestock houses was found. This was especially the case for the outside source in sows coarse PM, and turkeys fine PM; and the contribution of manure and skin in piglets coarse PM. Overall method accuracies varied from 52 to 88% showing classification rules could successfully distinguish more than 50% of particles and correctly assign them to its reference source based on cross-validation results.

Table 5.3. Average (Avg) percentage number contribution of the different PM sources to airborne fine PM (PM2.5) from different livestock species housing systems and accuracy of the classification. Standard error (SE) represents variation in the contribution between both surveyed livestock houses for the same housing system.

Sources	Broilers		Laying hens- floor		Laying hens-aviary		Turkeys		Piglets		Growing-finishing pigs		Dry and pregnant sows	
	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE
Feathers	30.1	20.7	38.4	22.9	10.5	5.8	27.3	19.1	-	-	-	-	-	-
Feed	8.1	8.1	3.0	1.8	2.4	2.4	1.7	1.7	15.9	5.1	3.7	1.5	14.5	2.2
Manure	14.0	7.3	49.5	22.0	84.7	1.0	8.9	8.9	73.9	1.6	88.8	1.3	69.8	2.4
Outside	28.8	9.1	9.2	0.8	2.4	2.4	44.3	37.1	7.0	5.3	5.4	0.1	4.1	4.1
Skin	-	-	-	-	-	-	-	-	3.2	1.4	2.1	0.1	11.7	0.6
Wood shavings	19.0	10.8	-	-	-	-	17.8	10.8	-	-	-	-	-	-
Accuracy (%)	73 - 86		73 - 74		52 - 75		67 - 83		57 - 79		78 - 84		74 - 75	

Table 5.4. Average (Avg) percentage number contribution of the different PM sources to airborne coarse PM (PM2.5-10) from different livestock species housing systems and accuracy of the classification. Standard error (SE) represents variation in the contribution between both surveyed livestock houses for the same housing system.

Sources	Broilers		Laying hens-floor		Laying hens-aviary		Turkeys		Piglets		Growing-finishing pigs		Dry and pregnant sows	
	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE
Feathers	35.1	13.1	12.8	1.9	8.9	2.7	32.4	17.6	-	-	-	-		-
Feed	8.2	1.8	2.5	1.5	2.5	0.0	3.7	1.7	14.1	7.0	5.0	0.7	3.2	3.2
Manure	29.8	7.2	83.6	1.5	86.7	4.7	40.7	8.0	41.3	34.1	71.0	0.4	58.7	25.4
Outside	16.5	4.5	1.0	0.0	1.9	1.9	13.7	7.9	11.6	9.8	10.8	6.6	34.1	32.5
Skin	-	-	-	-	-	-	-	-	33.0	31.3	13.1	5.5	4.0	4.0
Wood shavings	10.3	0.3	-	-	-	-	9.5	0.1	-	-	-	-	-	-
Accuracy (%)	76 - 85		78 - 88		75 - 84		62 - 76		74 - 79		78 - 81		63 - 80	

Using multiple linear regression

Results using multiple linear regression are shown in Table 5.5 (fine PM) and Table 5.6 (coarse PM), together with the variance explained by the regression model. Results showed higher contributions of manure to fine and coarse PM, and mostly lower contributions of feed and outside PM, compared with results when using classification rules based on decision trees. In piglets using multiple linear regression there was no estimated contribution of skin to number of collected particles, where manure particles composed the bulk of the collected PM in fine and coarse fractions.

Table 5.5. Average (Avg) percentage number contribution of the different PM sources to airborne fine PM (PM2.5) from different livestock species housing systems and variance explained by the regression model (R²). Standard error (SE) represents variation in the contribution between both surveyed livestock houses for the same housing system.

Sources	Broilers		Laying hens-floor		Laying hens-aviary		Turkeys		Piglets		Growing-finishing pigs		Dry and pregnant sows	
	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE
Feathers	28.4	21.5	4.4	1.1	16.0	8.7	43.2	15.3	-	-	-	-	-	-
Feed	0.0	0.0	9.6	9.6	0.0	0.0	0.0	0.0	2.4	2.4	0.4	0.4	0.7	0.7
Manure	67.7	18.2	74.2	1.8	84.0	8.7	22.9	12.8	91.2	4.0	98.3	1.7	78.9	4.1
Outside	0.3	0.3	11.8	8.9	0.0	0.0	0.2	0.2	6.4	6.4	1.0	1.0	0.4	0.4
Skin	-	-	-	-	-	-	-	-	0.0	0.0	0.4	0.4	20.0	4.4
Wood shavings	3.5	3.5		-	-	-	33.7	2.7	-	-	-	-	-	-
R²	79 - 82		49 - 87		94 - 96		88 - 97		43 - 74		78 - 96		71 - 78	

Table 5.6. Average (Avg) percentage number contribution of the different PM sources to airborne coarse PM (PM10-2.5) from different livestock species housing systems and variance explained by the regression model (R²). Standard error (SE) represents variation in the contribution between both surveyed livestock houses for the same housing system.

Sources	Broilers		Laying hens-floor		Laying hens-aviary		Turkeys		Piglets		Growing-finishing pigs		Dry and pregnant sows	
	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE
Feathers	17.2	6.8	6.3	6.3	10.2	9.9	31.7	3.2	-	-	-	-	-	-
Feed	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	6.0	6.0	4.2	4.2	0.0	0.0
Manure	82.8	6.8	93.7	6.3	87.7	7.8	35.8	1.5	94.0	6.0	84.5	1.8	52.1	33.4
Outside	0.0	0.0	0.0	0.0	2.2	2.2	0.0	0.0	0.0	0.0	0.0	0.0	40.7	40.7
Skin	-	-	-	-	-	-	-	-	0.0	0.0	11.3	2.4	7.3	7.3
Wood shavings	0.0	0.0	-	-	-	-	32.5	1.7	-	-	-	-	-	-
R²	86 - 97		88 - 88		95 - 96		86 - 94		44 - 61		76 - 88		51 - 85	

Overall, results indicated that in poultry, most of the PM originated from feathers and manure. Contribution of manure was again higher in coarse PM (ranging from 36 to 94%) compared with fine PM (ranging from 23 to 84%). Manure contribution was also higher in laying hen houses compared with broilers and turkeys; whereas feather contribution was higher in broilers and turkeys compared with laying hens. Wood shavings showed higher contributions in turkeys than in broilers, varying from 33 to 34% in fine and coarse PM in turkeys. In pigs, very high contributions of manure were found. The contribution of manure was again higher in fine PM (ranging from 79 to 98%) compared with coarse PM (ranging from 52 to 94%). Contribution of skin was lower (below 20%) compared with classification rules based on decisions trees, being highest in fine PM in sows and in coarse PM in growing-finishing pigs. Contribution of feed was estimated to be low (below 6%). It was higher in pigs compared with poultry, being the highest in piglets, in both fine and coarse PM. Contribution of the outside source was very low, except for sows in coarse PM. Standard errors of the estimated contributions between the surveyed livestock houses were generally low, except for the outside source in sows coarse PM. The variation explained by the model varied from 43 to 97%.

5.3.3.2. Contribution of sources to on-farm airborne PM expressed in mass

Applying equation 2, average mass per source in each livestock house was calculated. The contribution results presented in Table 5.3 to Table 5.6 were weighed by the average mass of each PM source in each livestock house to express percentage contribution of sources to on-farm airborne PM in mass.

Using classification rules based on decision trees

Results using classification rules based on decision trees shown in Table 5.7 (fine PM) and Table 5.8 (coarse PM) show different relative source contributions from number contributions. Although in poultry most of the number of particles originated from feathers and manure, the mass contribution of feathers decreased in broilers, but increased or did not vary in laying hens and turkeys, when expressed in mass. In mass, the contribution of manure was higher in laying hens compared with broilers and turkeys (same as for numbers), but also the contribution of feathers was higher in laying hens, especially compared with broilers. Although in pigs

most particles originated from manure, the mass contribution of skin considerably increased, in some cases more than ten-fold, ranging from 39 to 86% when expressed in mass, and thus decreasing the contribution of manure to below 46% in fine PM, and below 26% in coarse PM. Wood shavings showed approximately a two-fold increase in mass compared with number contributions, whereas the contribution of feed and outside was generally lower compared with number contributions.

Table 5.7. Average (Avg) percentage mass contribution of the different PM sources to airborne fine PM (PM2.5) from different livestock species housing systems using classification rules based on decision trees. Standard error (SE) represents variation in the contribution between both surveyed livestock houses for the same housing system.

Sources	Broilers		Laying hens-floor		Laying hens-aviary		Turkeys		Piglets		Growing-finishing pigs		Dry and pregnant sows	
	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE
Feathers	16.1	13.8	63.2	0.9	17.8	0.4	29.3	14.2	-	-	-	-	-	-
Feed	14.6	14.6	3.1	0.7	1.0	1.0	2.8	2.8	1.4	0.2	0.8	0.3	13.7	10.1
Manure	15.4	7.5	30.3	3.2	80.6	1.3	7.3	7.3	45.6	19.9	45.1	0.3	8.9	1.5
Outside	25.0	9.0	3.5	1.6	0.7	0.7	34.1	33.7	1.0	0.9	1.3	0.1	0.2	0.2
Skin	-	-	-	-	-	-	-	-	52.0	19.4	52.8	0.8	77.2	11.4
Wood shavings	28.8	15.8	-	-	-	-	26.6	15.0	-	-	-	-	-	-

Table 5.8. Average (Avg) percentage mass contribution of the different PM sources to airborne coarse PM (PM10-2.5) from different livestock species housing systems using classification rules based on decision trees. Standard error (SE) represents variation in the contribution between both surveyed livestock houses for the same housing system.

Sources	Broilers		Laying hens-floor		Laying hens-aviary		Turkeys		Piglets		Growing-finishing pigs		Dry and pregnant sows	
	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE
Feathers	8.0	1.0	32.6	3.6	27.0	9.0	46.1	28.5	-	-	-	-	-	-
Feed	2.3	1.3	3.4	2.1	1.1	0.8	1.6	1.4	1.8	1.7	0.3	0.1	0.3	0.3
Manure	51.0	30.2	63.1	4.8	68.7	11.4	35.5	21.7	20.2	20.1	11.7	4.4	25.5	3.8
Outside	3.1	1.6	1.0	0.8	3.2	3.2	4.1	3.4	1.1	0.2	2.0	1.2	35.6	35.1
Skin	-	-	-	-	-	-	-	-	77.0	21.5	86.0	5.7	38.6	38.6
Wood shavings	35.6	28.3	-	-	-	-	12.6	2.0	-	-	-	-	-	-

Using multiple linear regression

Results using multiple linear regression are shown in Table 5.9 (fine PM) and Table 5.10 (coarse PM). These results are comparable to using classification rules based on decision trees, showing similar trends and

differences when compared with number contributions, increasing the contribution of feathers in laying hens, of manure in broilers and turkeys, and of skin in pigs.

Table 5.9. Average (Avg) percentage mass contribution of the different PM sources to airborne fine PM (PM2.5) from different livestock species housing systems using classification rules based on multiple linear regression. Standard error (SE) represents variation in the contribution between both surveyed livestock houses for the same housing system.

Sources	Broilers		Laying hens- floor		Laying hens- aviary		Turkeys		Piglets		Growing- finishing pigs		Dry and pregnant sows	
	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE
Feathers	18.3	16.8	14.5	10.2	32.3	24.1	36.3	16.9	-	-	-	-	-	-
Feed	0.0	0.0	21.2	21.2	0.0	0.0	0.0	0.0	0.2	0.2	0.3	0.3	0.9	0.9
Manure	76.3	12.8	59.3	7.1	67.7	24.1	39.7	18.2	97.1	2.4	86.5	13.5	7.8	3.4
Outside	0.6	0.6	5.0	3.9	0.0	0.0	0.1	0.1	2.7	2.7	0.4	0.4	0.0	0.0
Skin	-	-	-	-	-	-	-	-	0.0	0.0	12.8	12.8	91.3	4.3
Wood shavings	4.7	4.7	-	-	-	-	23.8	1.3	-	-	-	-	-	-

Table 5.10. Average (Avg) percentage mass contribution of the different PM sources to airborne coarse PM (PM10-2.5) from different livestock species housing systems using classification rules based on multiple linear regression. Standard error (SE) represents variation in the contribution between both surveyed livestock houses for the same housing system.

Sources	Broilers		Laying hens- floor		Laying hens- aviary		Turkeys		Piglets		Growing- finishing pigs		Dry and pregnant sows	
	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE	Avg	SE
Feathers	3.4	1.3	12.1	12.1	26.9	26.1	22.0	2.5	-	-	-	-	-	-
Feed	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	4.3	4.3	0.3	0.3	0.0	0.0
Manure	96.6	1.3	87.9	12.1	72.8	25.8	58.0	1.7	95.7	4.3	14.1	2.0	14.7	1.3
Outside	-	-	0.0	0.0	0.3	0.4	0.0	0.0	0.0	0.0	0.0	0.0	42.0	42.0
Skin	-	-	-	-	-	-	-	-	0.0	0.0	85.7	2.3	43.3	43.3
Wood shavings	0.0	0.0	-	-	-	-	20.1	0.8	-	-	-	-	-	-

5.3.3.3. Comparison between methods

Results between classification rules based on decision trees and multiple linear regression in number of particles showed relatively high linear correlations ($R^2=0.75$ for fine PM and $R^2=0.61$ for coarse PM) (Figure 5.2). Correlations were higher for fine PM compared with coarse PM, probably influenced by the disagreement in the contribution of skin in piglets in coarse PM between methods.
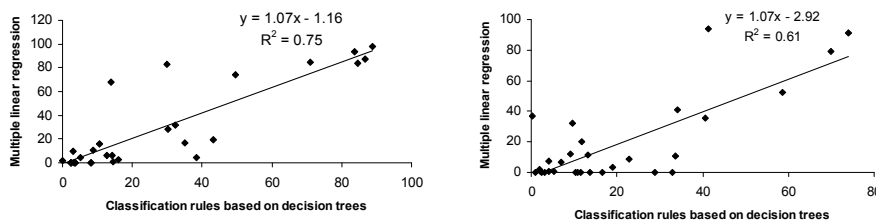
Figure 5.2. Linear relationship between classification rules based on decision trees and multiple linear regression source apportionment results for fine PM (left) and coarse PM (right).

5.4. Discussion

Based on particle numbers, feathers (ranging from 4 to 43% in fine and from 6 to 35% in coarse PM) and manure (ranging from 9 to 85% in fine and from 30 to 94% in coarse PM) were the most abundant in poultry. Manure (ranging from 70 to 98% in fine and from 41 to 94% in coarse PM) was the most abundant in pigs. Differences in source contributions between livestock species were mainly attributable to the different housing systems used and the presence of bedding, especially in broilers and turkeys. Morphology of the particles from the different sources could also explain such differences, for instance, the higher contribution of feathers in broilers and turkeys compared with laying hens. Broilers and turkeys feathers are generally lighter and looser, more "fluffy" in appearance, corresponding to plumules or down feathers, with shorter shafts than adult feathers as found in laying hens (Cambra-López et al., 2010b). Consequently, the nature of this type of feathers is probably more prone to become airborne.

The contribution of feed (which was below 16% in all cases) varied between livestock species, being constantly higher in pig houses compared with poultry houses. Perhaps the activity of the animals during feeding could explain such differences, being pigs generally more active during feeding time and thus creating more air movement because of their bigger body weights compared with poultry, and therefore probably contributing to more particles being generated, and becoming airborne, as explained in the processes and factors involved in PM being generated in livestock houses (Aarnink and Ellen, 2007). The type of feed and the feed processing could also play a role, as poultry feed is generally less crushed than pig feeds. The

contribution of outside particles was higher in pig houses compared with poultry, which were all tunnel ventilated except for turkeys (which also showed high outside PM contribution), compared with ceiling ventilation in pig houses.

Similar sources have been identified and similar number contributions have been reported in other studies. Donham *et al.* (1986) showed higher contributions of manure particles in the fine fraction of PM in pig houses as in this study. In poultry we found higher number of manure particles in coarse PM. The existence of two very distinctive morphological types of manure particles between poultry and pigs could be the cause of this difference (Cambra-López *et al.*, 2010b). Poultry excrete encapsulated uric acid crystals which are identified as round, smooth, spherical particles which can easily agglomerate, increasing in size. In pigs, however, this type of excretion does not exist, and manure particles are generally smaller, fragmented, rough, and angular particles, which are mostly found as individual particles falling into the fine range. The health implications of a higher number of manure particles found in the finer fractions in pig houses can be important; increasing the potential health risks associated with particle deposition ant its components in the deeper respiratory airways.

Feed, manure, pigs dander, mold, pollen and grains, insect parts, and mineral ash have been identified in PM samples from pig houses (Donham *et al.*, 1986). The contribution of feed to PM in livestock houses has been generally reported in higher ranges than those presented in this study. Heber *et al.* (1988) reported for finishing pigs, that most of PM originated from feed particles (about 65%) and to a lesser extent from manure and skin. Aarnink *et al.* (1999) also found higher contributions of feed in fattening pigs, but identified skin also as a major source. In poultry, Aarnink *et al.* (1999) obtained comparable results to those reported in this study, and identified down feathers and urine components as the most abundant in broilers. Feddes *et al.* (1992) found fecal material, mainly uric acid crystals as the main constituent in turkey houses. Fecal particles can resemble feed particles, furthermore, undigested feed components could be found in manure particles. The higher proportion of feed particles found in other studies, mainly starch in pig houses, could be attributable to the use of only light microscopy to distinguish between particles, and the higher content of

starch that can be found in pig's feces compared with poultry (Feddes *et al.*, 1992). Furthermore, total dust was used in these studies, as regards to fine and coarse segregated PM measurements as in our study. As reported by Feddes *et al.* (1992), the contribution of feed in particles bigger than 10 μm can be 30 times higher compared with the 0 to 5 μm size range.

The large differences in source contributions for a given housing system expressed as high standard errors could be part of the variation in the method used, because source apportionment models usually show high variations. Moreover, this could have been caused by the different housing conditions during samplings, together with the short sampling times used. Differences in PM concentrations between housing systems during sampling as in turkeys (Table 5.2) could also play a role in these differences between farms with the same housing system. The PM concentrations and emissions in a given livestock house can vary depending of the time of the day e.g. PM increases with feeding time and lighting periods (Calvet *et al.*, 2009; Hinz and Linke, 1998) and along a growing cycle e.g. with animal age, age of the bedding, or cleaning of the rooms (Hinz and Linke, 1998; Redwine *et al.*, 2002). Therefore, source contributions could vary depending on the activity moment within a day, but also between days; thus part of the between farm variation could be due to in-farm variation. More frequent measurements in the same livestock house through time could provide data to understand how PM source contributions can vary along a day and through a growing cycle.

Great variability between number and mass contributions results from the inherent variability of the morphological characteristics of PM (Cambra-López *et al.*, 2010c). Based on particle mass, feathers (ranging from 15 to 63% in fine and from 3 to 46% in coarse PM) and manure (ranging from 7 to 81% in fine and from 36 to 97% in coarse PM) were still the most abundant sources in poultry; whereas skin (ranging from 13 to 91% in fine and from 39 to 86% in coarse PM) was the most abundant in pigs. When estimating mass contributions it can be expected that bigger particles with bigger projected area diameters, although less numerous, gain relative importance. This is the case for wood shavings, and especially for skin particles. Differences amongst sources from different livestock species also result in different mass contributions of the sources. The different

morphological characteristics of (down) feathers from broilers compared with feathers from laying hens could explain why feathers in laying hens increase in relative contribution when expressed in mass compared with numbers. In broilers the opposite occurs. Other studies have reported similar source mass contributions. In growing-fattening pigs, Aarnink *et al.* (2004) reported high mass contributions of skin, comparable to those reported in this study. Our results suggest that probably hair from pigs, which shows big projected area diameters (Cambra-López *et al.*, 2010b), could also gain relative importance when expressed in mass contributions. However, in this study, hair was not included in the analysis because it showed very high carbon and oxygen peak in the SEM-EDX which was confused with the background filter composition.

In this study, it was necessary to assign an average shape (spherical) to a group of irregularly shaped particles. However, in practice, not all particles were spherical, but most of them were non-spherical, and it has been reported that the accuracy of sizing and weighing of particles using SEM can decrease when particles deviate from spheres (Willis *et al.*, 2002). This could be corrected for with the use of appropriate shape factors. The shape factor (S_v) is equal to 1 for spheres and varies depending on the shape of particles in relation to the resistance to a fluid motion. Noll *et al.* (1988) experimentally measured the shape factor of coarse atmospheric samples and determined a range from 1.35 to 3.15, but specific shape factors derived for biological or organic particles which could be used in our study are unknown. assuming spherical shapes to flat or flake-like particles could lead to overestimating mass of these particles because flake-shaped particles could be expected to have higher volume shape factors because it is difficult to keep the long axes of the particle in the direction of the moving flow (Zhang, 2004). Probably, these types of particles can offer more resistance to the flow. This could be the case of flattened skin particles in pigs as in our study. However, the purpose of this study was to give an insight into how the number contributions could vary when expressed in mass and not to provide an accurate estimation of particle weights derived from SEM observations.

Current European legislation sets limits to PM concentrations based on mass. Furthermore, particle size is critical to PM health and radiative effects.

Knowing how much and where PM will deposit in the respiratory system is important to assess health effects, because the smaller the size of PM, the deeper it can penetrate in the respiratory airways, compromising animal's and human's respiratory health (Donham, 2000; Radon *et al.*, 2001; Zuskin *et al.*, 1995). The relationship between particle mass and number contributions to PM in livestock houses discussed in this study can be also relevant to understand health risks associated to PM in livestock houses, and can be useful in designing reduction schemes. A mass-only approach to reduce PM would affect very little the number concentrations of the smaller particles found in the fine fraction. This fraction contains the fine and ultra-fine particles, with greater risks of adverse health effects because these particles can go beyond the larynx and penetrate into the unciliated respiratory system (EN, 1993). The control of particles bigger than 2.5 μm in diameter, however, is also relevant, because they can also cause adverse health effects in the upper respiratory airways. Furthermore, particles bigger than 2 μm in diameters have shown to contain high amounts of odorants (Cai *et al.*, 2006) and micro-organisms (Lee *et al.*, 2006). Both PM number and mass concentrations should be measured to tackle PM pollution related aspects within livestock houses, to develop reduction techniques and to assess their effects.

Overall, both methods used to quantify PM source contributions from livestock houses presented similar results and high levels of accuracy (expressed as overall correctly classified particles using classification rules, and as variance explained by the model using regression models). Therefore, using two independent methods, contribution results were consistent between them ($R^2=0.75$ for fine PM and $R^2=0.61$ for coarse PM). Differences between both methods, however, can be explained by: i) their own method characteristics, ii) by the use of different particle characteristics, and iii) by the discrepancies between single-particle chemical characteristics and average (bulk) elemental compositions. Furthermore, over and underestimation of source contributions when using classification rules based on decision trees can occur. Cambra-López *et al.* (2010a) determined overestimation of feathers (by an average factor of 1.6) and underestimation of wood shavings and outside source in poultry (by an

average factor of 0.65); and underestimation of outside source in coarse PM in pigs (by an average factor of 0.7).

In our results, differences in the obtained source contributions between classification rules based on decision trees and multiple linear regression were mainly caused by a higher contribution of manure when using multiple linear regression. This can be because manure is one of the most well-defined and homogeneous sources in terms of elemental composition compared with the rest (Cambra-López *et al.*, 2010a; Cambra-López *et al.*, 2010c). Moreover, multiple linear regression, which apportioned based on bulk particle chemical characteristics, searches for the combination that can predict better changes in the dependent variable in relation to changes in the independent variables using the least-squares method. The contribution of sources whose contributions were low using multiple linear regression (regression coefficients were very close to zero) could have been distributed amongst the manure source. Almeida *et al.* (2006) reported that with this method, the proportion of "unknown" fraction would be distributed amongst the identified sources with properties in common. Furthermore, when there are discrepancies between single-particle chemical characteristics and average elemental compositions, or when sources are not well-defined and are not chemically homogeneous, single-particle classification might apportion more accurately to these sources which show a more heterogeneous elemental composition than using average (bulk) elemental composition. According to Cambra-López *et al.* (2010a), this could be the case of feed and outside source, which show lower contributions when using multiple linear regression compared with classification rules based on decision trees in this study.

The differences found in piglets between methods could possibly be explained by the abundance of deposited, round, smoothed particles found in piglet houses in coarse PM (Figure 5.1e). These particles, which showed flattened surfaces and big sizes, could have been confused with skin particles when using classification rules which are based on particle morpho-chemical characteristics, but not when using only particle chemical characteristics as when using multiple linear regression, where skin did not show such high contributions. In any case, both methods require detailed and specific source profiles to apportion PM from livestock houses.

5.5.   Conclusions

1. Results presented in this study improve the understanding of where PM comes from in different livestock housing systems, not only in numbers but also in mass contributions. This can be valuable to choose the optimal dust reduction methods.

2. Using two independent methods, source apportionment results were consistent between classification rules based on decision trees and multiple linear regression ($R^2=0.75$ for fine PM and $R^2=0.61$ for coarse PM), and with detailed and specific chemical and morphological source profiles, both methods presented high levels of accuracy.

3. When there are high discrepancies between single-particle and average elemental composition or when sources are not well-defined and are not chemically homogeneous, using single-particle classification might apportion more accurately to these sources than using average (bulk) elemental composition.

4. Based on particle numbers, in poultry houses, most on-farm airborne PM originate from feathers (ranging from 4 to 43% in fine and from 6 to 35% in coarse PM) and manure (ranging from 9 to 85% in fine and from 30 to 94% in coarse PM). Manure contribution is higher in layer houses compared with broilers and turkeys; whereas feather contribution is higher in broilers and turkeys compared with laying hens. In broilers and turkeys, wood shavings contribute less than 34% of particle numbers.

5. Based on particle numbers, in pigs, most on-farm airborne PM originate from manure (ranging from 70 to 98% in fine and from 41 to 94% in coarse PM). Contribution of skin is below 33%, varying amongst pig categories.

6. The contribution of manure to on-farm airborne PM is higher in coarse PM in poultry, but higher in fine PM in pigs. We infer this to be due to the different morphological and thus airborne properties of individual manure particles from each species.

7. Feed has a negligible contribution to on-farm airborne PM compared with the rest of the sources, based on particle numbers. Its contribution, however, is higher in pigs compared with poultry.

8. When expressed in mass, big particles such as wood shavings, and especially skin gain relative importance compared with number of particles.

9. Based on particle mass, in poultry houses, still most on-farm airborne PM originate from feathers (ranging from 15 to 63% in fine and from 3 to 46% in coarse PM) and manure (ranging from 7 to 81% in fine and from 36 to 97% in coarse PM); but in pigs most on-farm airborne PM originate from skin (ranging from 13 to 91% in fine and from 39 to 86% in coarse PM).

5.6.   Acknowledgements

5.7.   References

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Chapter 6

Case-study: Ionization for reducing particulate matter emissions from poultry houses

Ionization for reducing particulate matter emissions from poultry houses

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Chapter 6

Abstract. We evaluated the effect of ionization in reducing particulate and gaseous emissions in broiler houses and its effect on particle size distribution. Furthermore we evaluated the performance of the tested ionization system and its influence on bird performance. The experiment was done during two consecutive rearing cycles in a pilot scale broiler house with four identical rooms. We measured concentrations of PM10 and PM2.5, airborne micro-organisms, ammonia, and odor of the incoming and outgoing air. Emissions were calculated by multiplying measured concentration difference of each pollutant by measured ventilation exchange rates. Performance of the system was evaluated through quantifying ion concentration, ozone production, and ultra fine particle concentration. Moreover, we recorded bird weight gain, consumption variables, mortality, and foot pad lesions. Overall measured mass emissions reductions were 36% for PM10, and 10% for PM2.5. Total mass was reduced less for PM2.5 because reduction efficiency decreased to the end of the growing period (P < 0.10). This coincided with increased particulate concentrations, increased ventilation exchange rates, and dust accumulation on surfaces. Higher reduction efficiencies were observed in relation to increased particle size. Ionization did not have a significant effect on micro-organism, ammonia or odor emissions; or on bird performance. Ionization proved to be a practical and effective technique for particulate reduction, with minimal maintenance to use in broiler houses. It is recommended to evaluate the use of ionization in commercial broiler houses to validate these results.

Keywords: Airborne micro-organisms, Ammonia, Broiler housing, Ionization, Particulate matter (dust).

6.1.   Introduction

Poultry houses, together with pig houses, show very high concentrations of air pollutants, mainly particulate matter (PM) (Takai *et al.*, 1998). In poultry, down feathers, mineral crystals from urine, and litter have been identified as major sources of PM (Aarnink *et al.*, 1999; Qi *et al.*, 1992). Broilers raised on litter, in particular, are key contributors to atmospheric PM emissions (Takai *et al.*, 1998).

Particulate matter is a potential hazard to health and welfare of humans and animals (Pope *et al.*, 2002). Several studies have reported increased respiratory problems in livestock farmers related to PM, such as chronic cough and/or phlegm, chronic bronchitis, allergic reactions and asthma-like symptoms (Andersen *et al.*, 2004; Donham *et al.*, 2000). Animal's respiratory health may also be compromised by PM (Collins and Algers, 1986; Donham and Leininger, 1984). Emitted PM from livestock houses is a concern because it can cause respiratory problems to people living in the vicinity of farms, as well (Lammel *et al.*, 2004; Radon *et al.*, 2002). Furthermore, atmospheric PM is relevant to climate change issues, such as cloud formation, radiative forcing, and it can contribute to atmospheric visibility impairment (IPCC, 2005).

Particulate matter in livestock houses differs from other types of PM because it is biologically active (Zhang, 2004). In livestock houses PM concentrations are 10 to 100 times higher compared with office and residential buildings. Particles can adsorb and carry toxic agents including irritating gases such as ammonia (NH_3) (Lee and Zhang, 2006; Takai *et al.*, 2002), volatile organic compounds and odor (Das *et al.*, 2004; Razote *et al.*, 2004), micro-organisms (Dennis and Gee, 1973; Seedorf *et al.*, 1998), bioactive components such as antibiotics (Hamscher *et al.*, 2003), and endotoxins (Schulze *et al.*, 2006). Attached to fine PM, these components are claimed to increase the potential health hazard of PM if they access the deeper respiratory airways (Donham and Leininger, 1984).

Reduction of PM emissions from livestock houses is a major challenge. Particulate matter which passes through a size-selective inlet with a 50 % efficiency cut-off at 10 µm aerodynamic diameter (PM10) are small enough to be inhaled and penetrate into the thorax. Particulate matter which passes

through a size-selective inlet with a 50 % efficiency cut-off at 2.5 µm aerodynamic diameter (PM2.5) can reach the alveoli in the lungs and have greater risks of adverse health effects (CEN-EN, 1993). Current legislation and European regulations (Integrated Prevention Pollution and Control-IPPC Directive 1996/61/EC, Council Directive 1999/30/EC, Directive 1996/62/EC, and Directive 2008/50/EC) require evaluation and control of PM10 and PM2.5 emissions to prevent harmful effects on people, and to protect the environment (Brunekreef and Maynard, 2008).

Consequently, there is growing pressure to find technically feasible and economically viable solutions to reduce PM emissions from intensive livestock houses. Air cleaning techniques in general, and air ionization in particular, have been identified to have a potential to reduce airborne PM concentration in different applications (Grabarczyk, 2001; Lee *et al.*, 2004; Niu *et al.*, 2001). Ionization is a technique which has been widely used to clean air in indoor environments. It is claimed to be more efficient in PM removal compared with conventional technologies (filtration and adsorption), requiring low energy costs, producing less hazardous reactants and by-products, and offering the potential of possible associated health benefits (Daniels, 2001).

The use of ions in air cleaning has been reported to reduce not only levels of particles by attachment to larger particles and agglomeration, but also of odors and volatile organic compounds in indoor air by oxidation (Daniels, 2007; Wu and Lee, 2004). Ions also have bactericidal effects and can reduce airborne micro-organisms (Krueger and Reed, 1976; Phillips *et al.*, 1964), and allergens (Dennis, 2003; Goodman and Hughes, 2004). Ionization has also been successfully used in control of infection caused by airborne pathogens both in humans and animals (Kerr *et al.*, 2006; Mitchell *et al.*, 2004; Seo *et al.*, 2001).

Ionization systems have been tested in poultry houses and hatching cabinets (Lyngtveit and Eduard, 1997; Mitchell *et al.*, 2000; Mitchell *et al.*, 2002; Mitchell *et al.*, 2004; Mitchell and Waltman, 2003; Quarantelli *et al.*, 2000; Richardson *et al.*, 2003; Ritz *et al.*, 2006). Some specific investigations have also been done in other livestock houses such as pig houses (Rosentrater,

2003; Tanaka and Zhang, 1996), cattle (Dolejs *et al.*, 2006), and rabbits (Chiumenti and Guercini, 1990).

Side effects related to air ionization are excessive electrostatic discharges and charging of objects, accumulation of precipitated dust on undesired surfaces in the room, reduced PM collection with increased thickness of dust layer on collection surfaces, and need of periodically cleaning of collection surfaces (Tanaka and Zhang, 1996). Some other side effects are the emission of ozone as a potential by-product of the ionization process (Boelter and Davidson, 1997; Britigan *et al.*, 2006), and the generation of submicron particles (Alshawa *et al.*, 2007).

An optimal design of an ionization system for use in livestock houses has not yet been fully developed, despite examples of research that have shown good results of ionization improving air quality in livestock houses (Rosentrater, 2004). In fact, the performance of ionization systems in different livestock housing applications still remains quite unpredictable, especially when dealing with particles from different size ranges, high PM concentrations, and increasing ventilation rates. Furthermore, the effect of air ionization on other pollutants such as airborne micro-organisms and gases needs to be examined.

The objective of this experiment was to evaluate the effect of air ionization in reducing PM (PM10 and PM2.5), airborne micro-organisms, NH_3 and odor emissions, and its effect on particle size distributions in broiler houses. Furthermore we evaluated the performance of the system in terms of ion concentration, ozone production, ultra fine particle concentration, and its influence on bird performance. Results will support a better understanding of the performance and dimensioning of ionization systems to use in broiler houses.

6.2. Material and methods

6.2.1. Experimental design

The effect of air ionization was studied in a pilot scale broiler house with four identical broiler rooms. Two of these rooms were randomly assigned to the ionization treatment, while the other two rooms served as control. The experiment was done during two consecutive rearing cycles.

6.2.2. Housing and animals

The broiler house was located at the experimental station 'Het Spelderholt' in Lelystad, The Netherlands. Each room measured 8.3 x 16.0 m (133.6 m²) and had a volume of 500 m³. Each room had four feeding lines with seven feeders, and eight drinking lines with 180 drinking nipples in total. The rooms were heated by a central heating system with heaters on the side walls underneath the air inlet openings. Rooms were mechanically ventilated, and each room had three ventilators suspended from the ceiling, of which one was continuously working, and the other two working when needed. The three ventilators in each room had a total maximum capacity of 21,000 m³ h^{-1}.

At the start of the experiment, 2676 one-day old birds, a mixture of males and females, were placed in each room at a density of 20 birds m^{-2}. Wood shavings were used as litter, spread at a density of approximately 1 kg m^{-2} in each room. Broilers were delivered to the slaughter house at an age of 35 days, at a target weight of approximately 2000 g. Broilers had free access to feed and drinking water. The first 10 days, broilers received a starter diet (mash), followed by a grower diet (days 11-28, pellet), and a finisher diet (days 29-35, pellet). Broilers received all necessary vaccinations.

During the first two days, rooms were continuously lighted. During the rest of the rearing cycle, an intermittent light scheme was given of 8 h light and 4 h dark (07:45 – 15:45 (light); 15:45 – 19:45 (dark); 19:45 – 03:45 (light); 03:45 – 07:45 (dark)). Light intensity was the same for each room (20 lux). Inside climate was also controlled the same for each room. Temperature was gradually decreased from 33°C on day one of the rearing cycle, to 20°C on day 35. Minimum ventilation was controlled at 1 m³ kg^{-1} live weight.

6.2.3. Ionization system

The ionization system "Electrostatic Particle Ionization" (EPI) system (Baumgartner Environics, Inc., USDA Patent #6,126,722, U.S.) was used. The EPI system consisted of two rows of inline, negative DC ionization units running along the length of each ionization room. Ionization units were composed of a discharge electrode (ion generator) and a grounded collection plate. These units were installed by the manufacturing company, suspended at a height of approximately 2.5 m above the litter. The discharge

electrode consisted of a conductive tube with sharp pointed electrodes at 2.54 cm intervals, pointing towards the litter. This electrode was connected to a high voltage power supply (EPI, Baumgartner Environics, Inc., Olivia, U.S.) to create a high density electron array (-30 kV DC), limited to a current of 0.9 mA to assure safety.

The grounded collection plate consisted of two steel plates during the first rearing cycle. These two plates plus four aluminum sheets were used during the second rearing cycle, to improve collection efficiency. Grounded collection plates were located close to the discharge electrodes to maximize the electron output. Emitted electrons generated negatively charged ions. These ions charged circulating airborne particles, which were consequently attracted by the grounded plates and room surfaces. Cleaning of plates and sheets was modified between first and second rearing cycle: grounded collection plates were manually shaken and cleaned every other day in the first rearing cycle, and the aluminum sheets were mechanically brushed off in the second rearing cycle. Each aluminum sheet had a pair of brushes attached to it, which were pulled along the length of the plate for wiping off dust. Collected dust fell into a plastic bag, one for each sheet. Cleaning frequency in the second rearing cycle increased in time, from once a week in the first week, to twice a week from week 2 to week 4, to daily in week 5. The lay-out of the EPI system used in the first and second rearing cycle is shown in Figure 6.1.

6.2.4. Measurements

6.2.4.1. Particulate matter (PM10 and PM2.5)

Concentrations of PM10 and PM2.5 of the exhaust air were weekly measured, starting in the second week of each rearing cycle. Particulate matter concentrations were gravimetrically measured. These data were used to calculate emissions. Particulate matter was sampled during 24 h (from noon to noon) with cyclone pre-separators (URG corp., Chapel Hill, U.S.) for PM10 (following CEN-EN 12341, CEN, 1998) and PM2.5 (following CEN-EN 14907, CEN, 2005), and the aimed particle sizes were collected on glass fiber filters (Ø 47 mm, type GF-3, Macherey-Nagel, Düren, Germany).
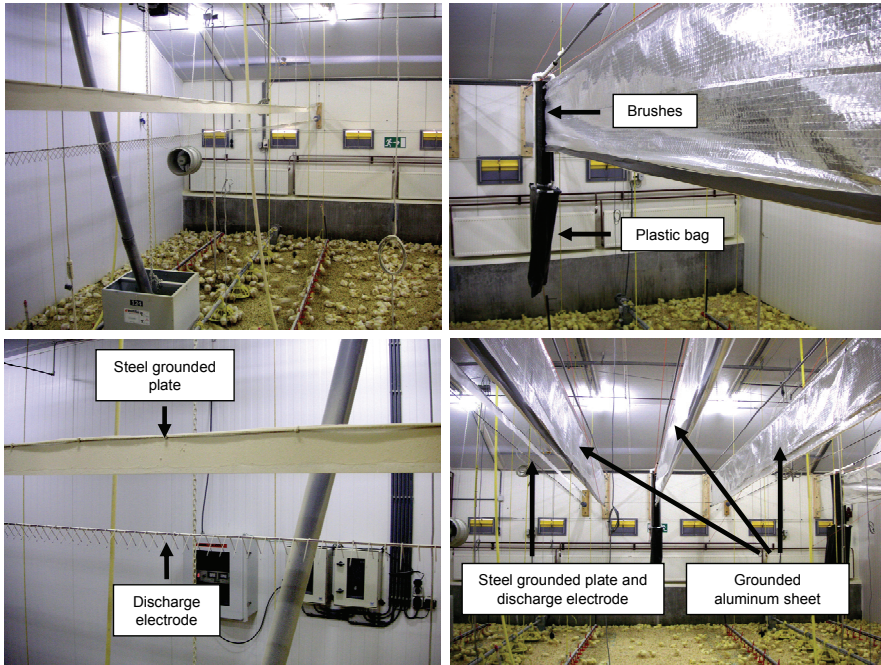
Figure 6.1. Pictures of the EPI system installed in the ionization rooms in the broiler house. Picture on the left: suspended ionization unit above litter (top), and detail of discharge electrode and steel grounded plate during the first rearing cycle (bottom). Picture on the right: suspended aluminum grounded sheets with brushes for mechanical cleaning and plastic bag for dust collection (top), and general view of both steel plates and aluminum sheets during the second rearing cycle (bottom).

Sampling position was close to the inlet of the ventilation shafts, at a height of 3 m. The inlet of the cyclones was placed at a horizontal distance of 0.5 m from the border of the exhaust opening, and at a vertical distance of 0.10 m underneath the exhaust opening. One PM10 cyclone and one PM2.5 cyclone were placed outside the broiler house to measure background PM concentrations, as well.

Sampled air was drawn into the sampler at an air flow rate of 16.7 L min^{-1}, using stationary pumps (Charlie HV, Ravebo Supply B.V., Brielle, The Netherlands). Pumps were able to keep a constant air flow using a temperature sensor at the same position as the inlet of the cyclone PM collector. This flow could even be kept constant when the PM sampling filter was heavily loaded. The volume of air passing through the cyclones was measured by a gas meter within the pump and corrected for the temperature measured at the sampling point.

The unloaded glass fiber filters were stabilized for 48 h under standard conditions: temperature 20°C ± 1°C, and 50% ± 5% relative humidity. Each filter was then weighed four times using a Mettler balance (minimum reading 10 μg), according to CEN-EN 14907 (CEN, 2005). The average value was calculated as the filter weight. For the loaded filters, the same weighing procedure was adopted. The weight difference between loaded and unloaded filters equaled the amount of collected PM. For a detailed description of the sampling procedure see Zhao *et al.* (2009).

Weekly PM10 concentrations were also continuously measured in the exhaust air with a light scattering system (DustTrak TM Aerosol Monitor, model 8520, TSI Incorporated, Shoreview, U.S.). One DustTrak per room was used on the same days and at the same distance and position from the ventilator as the cyclones. DustTraks were also used to determine PM10 concentrations in the exhaust air during the first two weeks of each rearing cycle, before the start of cyclone measurements.

Incidental measurements were carried out during cleaning of the ionization system. Once in the first rearing cycle, one DustTrak per room was left recording for 72 h, starting 24 h before cleaning. This incidental measurement was done during days 31 to 33 in the first rearing cycle.

6.2.4.2. Particle size distribution

Particle counts (number concentrations) in the different size ranges were measured in each room using an optical particle counter (OPC, model 1.109, Grimm Aerosol Technik GmbH & Co., Ainring, Germany). Size distribution of PM was determined for sizes between 0.25 and 32 μm (optical latex equivalent diameter), classified in 31 channel sizes. Sampling air flow rate was 1.2 L min^{-1}. The equipment was set to a sampling interval of one minute. Each room was sampled during 30 min, twice during the first rearing cycle (days 33 and 34), and four times during the second rearing cycle (days 5, 19, 26 and 34). Average values for the different size ranges were calculated.

6.2.4.3. Personal PM10 sampling

Personal dust load (PM10) was determined by hanging a DustTrak close to the worker's breathing zone, when simulating daily animal care routine

activities between 09:00 a.m. to 11:00 a.m. The sampler was attached to the person's lapel, at a height of approximately 1.5 m, with the PM10 inlet facing upwards and close to the nose and mouth. Workers wore masks to prevent directly breathing into the inlet. Each room was randomly sampled for 7 min, three times during the first rearing cycle (days 25, 32 and 33), and twice during the second rearing cycle (days 25 and 32). One minute values were stored for average PM10 concentrations in mg m^{-3}.

6.2.4.4. Ammonia

Ammonia concentrations in the inlet and exhaust air were continuously sampled in each rearing cycle with a NO_x monitor (model ML8840, Monitor Labs, Englewood, U.S.). Air was sampled in each room, at the exhaust of the middle ventilator which was working continuously. Samples of air inlet were also taken outdoors. Air was conducted through heated teflon tubes to the converter. In the converter, NH_3 present in the air was converted into nitrogen oxide (NO) at 775°C. From the converter, air was transported through heated tubes to the NO_x monitor where NO concentrations were measured and recorded continuously. The monitor was weekly calibrated with a gas of 40 ppm NO in nitrogen (N_2) and the flow was checked. Hourly average values were recorded and used to calculate NH_3 emissions.

6.2.4.5. Airborne micro-organisms

The impingement method was used to determine total airborne bacteria, and fungi and mold populations in each room. Samples were taken weekly starting on the second week of each rearing cycle, at the same location as PM sampling, at a 0.5 m horizontal distance from the border of the exhaust opening and at a vertical distance of 0.10 m underneath the exhaust opening. Samples were taken during 15 min.

Duplicate autoclaved all-glass impingers with 30-mm jet-to-bottom spacing (AGI-30, All Glass Impinger, Ace Glass Incorporated, Vineland, U.S.) were used in each room. Sampled air was drawn into the impinger at a calibrated air flow rate of 12.5 L min^{-1}, using stationary pumps (Charlie HV, Ravebo Supply B.V., Brielle, The Netherlands). Impingers were used with 20 mL of sterile 1% peptone-distilled water with 0.005% defoamer (Winterhalter Gastronom, GmbH). After sampling, samples were transported to the

laboratory at a temperature between 4ºC and 8ºC, and analyzed within 8 h. The final volume was measured and corrected for evaporation. Samples were serial 10-fold diluted in 0.1% peptone-distilled water, and 0.1 mL samples were plated onto duplicate plates: Plate Count Agar for total bacteria, and Oxotetracycline-Gentamicine-Glucose-Agar for total fungi and mold. Plates were then incubated at 30°C for 3 days, for total bacteria; and at 25°C for 3 to 5 days, for total fungi and mold. Colony forming units (cfu) were counted on plates containing between 30 and 300 colonies (Thorne *et al.*, 1992). Airborne concentrations of total bacteria, and total fungi and mold were determined by multiplying the cfu by the dilution volume, and divided by the volume plated (0.1 mL). Colony forming units were then calculated for the volume of sampled air, sampling time, and flow rate. These data were used to calculate airborne micro-organism emissions.

6.2.4.6. Odor

Odor concentrations were measured twice in each room at the same days for both rearing cycles: at day 24 and at day 31. Two-hour odor samples were collected using the "lung principle". A new 40 L Nalophan odor sampling bag was placed in a rigid container. The bags had been previously flushed with compressed and odorless air three times. During sampling, air was removed from the container with a vacuum pump, and the vacuum in the container caused the bag to fill with a volume of air equal to the volume of air removed from the container. Flow rate of air entering the sample bag was 0.5 L min^{-1}. Odor samples were transported and stored according to CEN-EN standard 13725 (CEN, 2003). Odor concentrations were determined by olfactometry within 30 h after sampling (CEN, 2003). These data were used to calculate odor emissions.

6.2.4.7. Ventilation rate

Ventilation rate was measured with calibrated anemometers (ATM.56, Fancom, Panningen, The Netherlands) with the same diameter as the ventilation shafts, one in each of the three ventilation shafts of each room. Hourly averages were stored in a data logging system.

6.2.4.8. Environmental parameters

Temperature and relative humidity of the exhaust air were continuously measured in each room with combined sensors (Rotronic Instrument Corp., U.S.). Hourly means were stored in a data logging system. Outside temperature and relative humidity were recorded in the same way.

6.2.4.9. Performance of ionization system

Ion concentration

Ion concentrations were measured in each ionization room with an air ion counter (AlphaLab Inc., U.S.), with a range from 199.9 to 1000 positive or negative 10^6 ions cm^{-3}. Ion concentrations were measured weekly at five locations along the width of each ionization room. The air ion counter sampled air and deposited ions onto an internal collector plate. The number of elementary charges on the collector plate was determined by voltage measurement.

Ozone concentration

Ozone concentrations were measured in each ionization room with ozone detector tubes (Kitagawa, No. 182U, Hatech Gasdetectietechniek, The Netherlands). Samples were taken inside each room, at a height of 1.5 m at the center of the room with a manual pump. These tubes were selected for being the most sensitive. Lowest detection limit of the tubes (0.01 ppm) could be achieved by increasing number of pump strokes. Two measurements per week were carried out during the first two weeks because of the low ventilation exchange rates, followed by weekly measurements during the rest of both rearing cycles.

Ultra fine particle number concentration

Ultra fine particle number concentrations were determined using a condensation particle counter (CPC, Series 5.400, Model number 5.403, Grimm Aerosol Technik GmbH & Co., Ainring, Germany) which determined particle number concentrations with a higher cut-off diameter of 1100 nanometers (nm), and a lower cut-off diameter of 5 nm. This instrument measured particle concentrations up to 10^7 particles cm^{-3} with a time resolution of one second. Ultra fine particle concentrations were

measured twice per room during the first rearing cycle (days 33 and 34), and three times per room during the second rearing cycle (days 19, 26 and 34). Each room was sampled during 30 min. The equipment was set to store one minute averages. From these data mean 30 min values were calculated.

Bird performance

Broilers were weighed on arrival at the broiler house and before transport to the processing plant on day 35 of each rearing cycle to determine the start and end weights. All birds were weighed in one group per room. Furthermore, a sample of 100 broilers (50 male, 50 female) per room were weighed on day 21 and 34. Total feed intake was determined on day 21 and 35, and feed conversion ratios were calculated, corrected for mortality. Mortality numbers and weights of these broilers were recorded daily per room. Water consumption was recorded daily.

Bird foot pad lesions were evaluated on day 33 of each rearing cycle. Before transport to the processing plant, the quality of the exterior of the broilers was scored in a random sample of 50 male and 50 female broilers. Birds were scored on breast dirtiness, breast irritations, scabby hips (thigh scratches), and hock burns as described in Van Harn (2008). Foot pad lesions were scored according to the protocol described by Berg (1998).

A summary of the measurement techniques used during the whole experiment (both rearing cycles) is presented in Table 6.1. A schematic diagram of a broiler room showing sampling locations with respect to ionization system and the ventilator exhaust is given in Figure 6.2, as an example. Sampling locations were the same in control rooms.

Table 6.1. Summary of evaluated parameters, measurement methods, units, sampling duration, and frequency of measurements for both rearing cycles

Parameter	Measurement method	Unit	Sampling duration	Frequency of measurement (days in the rearing cycle)[*]
Particulate matter				
Cumulative PM10 and PM2.5	Gravimetry	mg m^{-3}	24 h	9, 16, 23, 30 and 33
Continuous PM10	Nephelometry (light scattering)	mg m^{-3}	24 h	2, 9, 16, 23, 30 and 33
Incidental PM10	Nephelometry (light scattering)	mg m^{-3}	72 h	31 to 33 (1st rearing cycle)
Particle size distribution	Laser spectrometry (90° light scattering)	particles cm^{-3}	30 min	33 and 34 (1st rearing cycle) 5, 19, 26 and 34 (2nd rearing cycle)
Personal PM10	Nephelometry (light scattering)	mg m^{-3}	7 min	25, 32 and 33 (1st rearing cycle) 25 and 32 (2nd rearing cycle)
Ammonia	NH$_3$-analyzer and NO$_x$-converter	mg m^{-3}	Continuous	
Airborne micro-organisms				
Total bacteria, fungi and mold	Liquid impingement	log cfu m^{-3}	15 min	15, 22, 29 and 32
Odor	Dynamic olfactometry	ou$_E$ m^{-3}	2 h	24 and 31
Ventilation and environmental parameters				
Ventilation rate	Anemometry	m^3 h^{-1}	Continuous	
Temperature	Temperature sensor	°C	Continuous	
Relative humidity	Relative humidity sensor	%	Continuous	
Performance ionization system				
Ion concentration	Ion meter	ions m^{-3}	1- 2 min	1, 7, 14, 21 and 28
Ozone concentration	Colorimetric chemical reaction	ppm	6 min	1, 4, 7, 14, 21 and 28
Ultra fine particle number	Particle condensation and laser spectrometry	particles cm^{-3}	30 min	33 and 34 (1st rearing cycle) 19, 26 and 34 (2nd rearing cycle)
Bird performance				
Bird weight	Weighing	g	-	0, 21, 34 and 35
Feed consumption	Weighing	g day^{-1} bird^{-1}	-	0, 21 and 35
Water consumption	Water meter	mL day^{-1} bird^{-1}	-	Daily
Mortality	Visual observations	%	-	Daily
Foot pad lesions	Visual observations - Scoring	Score	-	33

[*] Frequency of measurements is the same for both rearing cycles, otherwise stated in the table
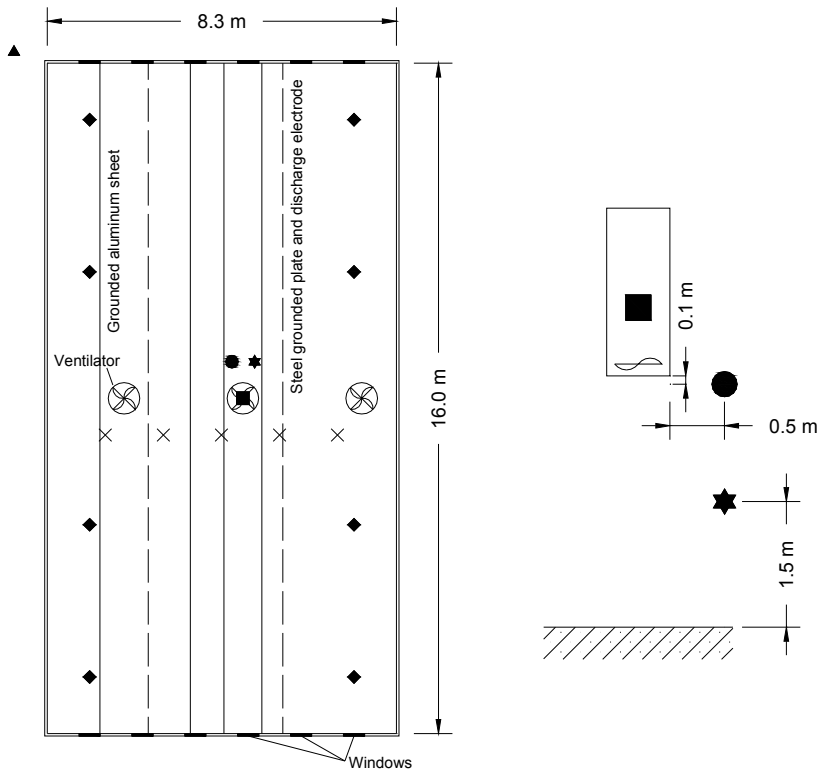
Figure 6.2. Schematic diagram of an ionization room showing sampling locations and position of ionization system, and sampling positions with respect to ventilation shaft inside the room (●PM and micro-organisms;✱ Particles size distribution, ultra fine particles, and ozone; ■NH₃ and ventilation rate; ◆Temperature and relative humidity;✕Ion concentration;▲Outdoor PM, NH₃, temperature, and relative humidity).

6.2.5.   Data analysis

6.2.5.1. Estimation of pollutant emission rates

To calculate PM (PM10 and PM2.5), airborne micro-organisms, NH₃, and odor emissions, the concentration measured outdoor (inlet) was subtracted from the concentration measured indoor (exhaust), and multiplied with the ventilation exchange rate following equation 1:

$$Emission = (C_{exhaust} - C_{inlet}) \times Q \tag{1}$$

where:

$C_{exhaust}$= concentration of specific pollutant i in the exhaust air of the room

C_{inlet}= concentration of specific pollutant i in the inlet of the room

Q= ventilation exchange rate (m^3 h^{-1})

6.2.5.2. Estimation of reduction efficiency

Emission data were used to calculate reduction percentages, expressed as the relative difference between control and ionization rooms for each tested variable.

6.2.5.3. Statistical analysis

Particulate matter emissions were statistically analyzed assuming a linear relationship at log-scale between PM emissions and time, with intercept β_0, and slope β_1 (eq. 2) using Genstat (Genstat Committee, 2008). Natural log-transformed data were used to determine the effect of ionization on PM10 and PM2.5 emissions. Reductions for a given PM fraction were calculated as the relative difference in PM emissions between ionization and control rooms. The linear relationship between PM emissions and time is as follows:

$$Log\left(\underline{Y}_{ijk}\right)= \left\{\beta_{0i} + \underline{\varepsilon}_{0ij}\right\}+ \left\{\beta_{1i} + \underline{\varepsilon}_{1ij}\right\}\times t + \underline{\varepsilon}_{ijk} \qquad (2)$$

where:

$\underline{Y}_{ijk}$ = response variable (PM10 and PM2.5 emissions) of measurement k in room j for treatment i

t= day within rearing cycle

β_{0i}= intercept i (t=0, at the start of the rearing cycle)

β_{1i}= linear increasing trend in response during the rearing cycle of treatment i

$\underline{\varepsilon}_{0ij} \sim N(0,\sigma^2_{0ij}), \varepsilon_{1ij} \sim N(0,\sigma^2_{1ij})$ = random room effect of intercept and increasing trend, respectively, within treatment i.

$\underline{\varepsilon}_{ijk} \sim N\left(0;\sum\tau_k,\phi_k\right)$ = random day effects correlated within room (auto-regression), variances can differ between different measuring days.

Statistical significant differences between control and ionization rooms for PM concentrations and emissions, micro-organisms, NH_3, and odor

emissions were determined with a two-tailed t-test for one treatment with two levels (ionization and control) using Genstat (Genstat Committee, 2008). Differences with P values less than 0.05 were considered to be statistically significant, assuming equal variance of groups. The t-test was also applied for particle counts per size range. Average values per room within rearing cycles were the experimental units in the t-test analyses.

Effects of cleaning and lighting on PM10 reductions were analyzed with one-way ANOVA using Genstat (Genstat Committee, 2008) with ionization, control, cleaning, lighting, and its interactions as sources of variance. Average PM10 concentration values per treatment collected over 72 h in the first rearing cycle were the experimental unit in this ANOVA analyses. Also, effects of cleaning on PM10 concentrations in ionization rooms were analyzed with one-way ANOVA using Genstat (Genstat Committee, 2008) with cleaning as source of variance. Average PM10 concentration values in ionization rooms, before and after cleaning, collected over 72 h in the first rearing cycle were the experimental unit in this ANOVA analyses.

Results of bird performance and foot pad lesions were statistically analyzed with one-way ANOVA using Genstat (Genstat Committee, 2008), with ionization and control as sources of variance. Average values per room within rearing cycles were the experimental units in the ANOVA analyses.

6.3. Results

6.3.1. Ventilation rates and environmental parameters

Average ventilation exchange rates during the experiment (1.8 m^{-3} h^{-1} $bird^{-1}$) were similar for both rearing cycles and increased along the rearing cycle from minimum ventilation exchange rates of 0.01 m^{-3} h^{-1} $bird^{-1}$, to maximum ventilations of approximately 5 m^{-3} h^{-1} $bird^{-1}$ (Figure 6.3). Daily variations in ventilation exchange rates were observed, determining daily emission rate patterns. During the second rearing cycle, however, average ventilation exchange rates were approximately 10% lower compared with the first rearing cycle, because a decrease of outside temperatures was registered after day 23 of the second rearing cycle.

Temperature and relative humidity were similar in all rooms during the whole experiment. Outside temperature was on average 16.6°C during the first rearing cycle and 16.1°C during the second rearing cycle. Overall average indoor temperatures were approximately 25°C for all rooms. Relative humidity varied from 56% to 68% for all rooms.

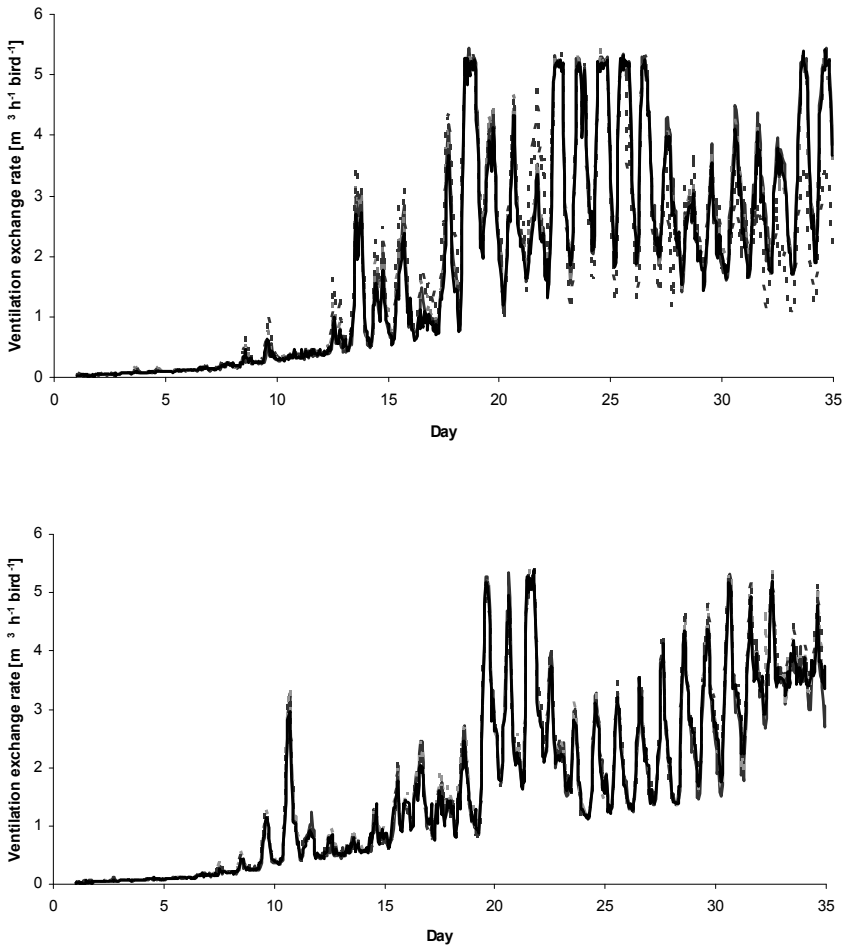Figure 6.3. Average daily ventilation exchange rate (m³ h⁻¹ day⁻¹) along the rearing cycle for the first rearing cycle (top), and for the second rearing cycle (bottom). Dark continuous lines show control and dashed grey lines show ionization rooms.

6.3.2. Particulate matter (PM10 and PM2.5) concentrations and emissions

Table 6.2 provides average PM10 concentrations at the exhaust, and emissions for the whole experiment (both rearing cycles), for ionization and control rooms. Inlet PM10 concentrations were below 0.051 mg m^{-3} during both rearing cycles, on average (standard deviation, SD) 0,023 (0,015) mg m^{-3}. Concentrations of PM10 increased with age of the birds in ionization and control rooms, and so did emissions. Emission rates increased with ventilation exchange rates and also with increasing PM concentrations indoors. Concentrations and emissions in ionization and control rooms were lower in the first rearing cycle compared with the second. Total average (SD) PM10 concentrations were 1.267 (0.720) mg m^{-3} during the first rearing cycle; 0.744 (0.262) mg m^{-3} during the second rearing cycle. Total average (SD) PM10 emissions were 90.49 (69.97) mg bird^{-1} day^{-1} during the first rearing cycle; 47.04 (24.44) mg bird^{-1} day^{-1} during the second rearing cycle. For both rearing cycles, concentrations and emissions of PM10 in ionization rooms were lower ($P < 0.05$) compared with control rooms (Table 6.2).

Table 6.2. Average PM10 concentrations at the exhaust (mg m^{-3}), emissions (mg bird^{-1} day^{-1}), and standard deviation (SD) for both rearing cycles (normal scale-measured values).

	Treatment	PM10 concentration (mg m^{-3})	SD	PM10 emission (mg bird^{-1} day^{-1})	SD
Day 2*	Control	0.241[a]	0.066	0.365[a]	0.110
	Ionization	0.054[b]	0.043	0.078[b]	0.060
Day 9*	Control	0.544[a]	0.221	6.528[a]	3.996
	Ionization	0.126[b]	0.020	1.688[a]	0.371
Day 16	Control	0.739[a]	0.215	20.392[a]	4.771
	Ionization	0.410[b]	0.029	13.130[b]	0.926
Day 23	Control	0.988[a]	0.273	59.784[a]	11.469
	Ionization	0.586[b]	0.051	36.770[b]	11.296
Day 30	Control	1.494[a]	0.571	107.773[a]	30.437
	Ionization	0.998[a]	0.357	70.654[a]	16.605
Day 33	Control	1.620[a]	1.018	146.907[a]	96.249
	Ionization	1.208[a]	0.612	94.723[a]	44.234
Total PM10	Control	1.210[a]	0.660	83.714[a]	67.157
(average from days 16 to 33)	Ionization	0.800[b]	0.457	53.820[a]	38.896

* Days 2 and 9 correspond to 24 h average DustTrak values

[a,b] Means within a day with different superscripts differ significantly ($P < 0.05$)

On a logarithmic scale, PM10 emissions were on average lower ($P < 0.001$) in ionization rooms compared with control rooms, given by intercept β_0 in the linear relationship shown in equation 2. Reduction was not influenced by the age of the birds (from day 16 onwards) ($P > 0.05$), given by slope β_1 in the linear relationship shown in equation 2. Overall measured mass reduction for PM10 emissions (at normal scale) was on average (SD) 36% (2%) lower in ionization rooms compared with control rooms. Measurements done in the first two weeks with DustTraks showed higher emission reductions (77%).

Table 6.3 provides average PM2.5 concentrations at the exhaust, and emissions for the whole experiment, for ionization and control rooms. Inlet PM2.5 concentrations were below 0.026 mg m^{-3} during both rearing cycles, on average (SD) 0.016 (0.007) mg m^{-3}. Concentrations of PM2.5 were lower than concentrations of PM10. The PM2.5 was on average 15% of PM10 concentration. Concentrations of PM2.5 also showed an increase with age of the birds, and so did emissions, although less pronounced compared with PM10. Concentrations and emissions in ionization and control rooms were lower in the first rearing cycle compared with the second. Total average (SD) PM2.5 concentrations were 0.088 (0.054) mg m^{-3} during the first rearing cycle; 0.044 (0.016) mg m^{-3} during the second rearing cycle. Total average (SD) PM2.5 emissions were 4.79 (4.67) mg bird^{-1} day^{-1} during the first rearing cycle; 2.32 (1.47) mg bird^{-1} day^{-1} during the second rearing cycle. For both rearing cycles, no significant differences in PM2.5 concentrations in ionization compared with control rooms was observed. For both rearing cycles, only on day 30, emissions of PM2.5 were significantly lower ($P < 0.05$) in ionization rooms compared with control rooms (Table 6.3).

On a logarithmic scale PM2.5 emissions were on average lower ($P < 0.05$) in ionization rooms compared with control rooms, given by intercept β_0 in the linear relationship shown in equation 2. There was a tendency ($P < 0.10$) for an effect of day number on the reduction of the ionization system, given by slope β_1 in the linear relationship shown in equation 2. Overall measured mass reduction for PM2.5 emissions (at normal scale) was on average (SD) only 10% (33%) lower in ionization rooms compared with control rooms. Average calculated reductions were 67% (day 16), 27% (day 23), 28% (day 30), and -15% (day 33).That no effect was measured on day 33 (Table 6.3)

was mainly caused by a negative reduction of -30% in the first rearing cycle on day 33.

Table 6.3. Average PM2.5 concentrations at the exhaust (mg m^{-3}), emissions (mg bird^{-1} day^{-1}) and standard deviation (SD) for both rearing cycles (normal scale-measured values).

	Treatment	PM2.5 concentration (mg m^{-3})	SD	PM2.5 emission (mg bird^{-1} day^{-1})	SD
Day 16	Control	0.050[a]	0.037	0.944[a]	0.699
	Ionization	0.023[a]	0.008	0.311[a]	0.148
Day 23	Control	0.050[a]	0.011	2.087[a]	0.983
	Ionization	0.041[a]	0.007	1.525[a]	0.389
Day 30	Control	0.084[a]	0.019	4.817[a]	0.623
	Ionization	0.066[a]	0.016	3.474[b]	0.598
Day 33	Control	0.094[a]	0.044	7.106[a]	3.679
	Ionization	0.120[a]	0.087	8.172[a]	6.447
Total PM2.5	Control	0.069[a]	0.034	3.738[a]	3.036
(average all days)	Ionization	0.062[a]	0.055	3.369[a]	4.239

[a,b] Means within a day with different superscripts differ significantly ($P < 0.05$)

Figure 6.4 shows continuous 72 h measurements using DustTraks. During these 72 h measurements, PM10 concentrations (SD) were 27% (22%) lower ($P < 0.001$) in ionization rooms compared with control rooms (Figure 6.4). Two high and two low PM10 concentration periods in ionization and control rooms during 24 h were identified (Figure 6.4). Concentrations of PM10 increased when lights were on (light periods), and decreased when lights were off (dark periods), showing sharp PM10 spikes coinciding with lights on (07:45 and 19:45), and increased animal activity. PM10 concentrations (SD) were 27% (6%) lower ($P < 0.001$) in ionization rooms compared with control rooms during light periods, and 33% (7%) lower ($P < 0.001$) during dark periods.

Cleaning ionization grounded electrodes showed no statistically significant difference in PM10 concentrations in ionization rooms before and after cleaning, despite reductions of PM10 concentrations increased by 10%, 24 h after cleaning (Figure 6.4). Only lighting schedule showed a significant effect on PM10 concentrations in ionization rooms.
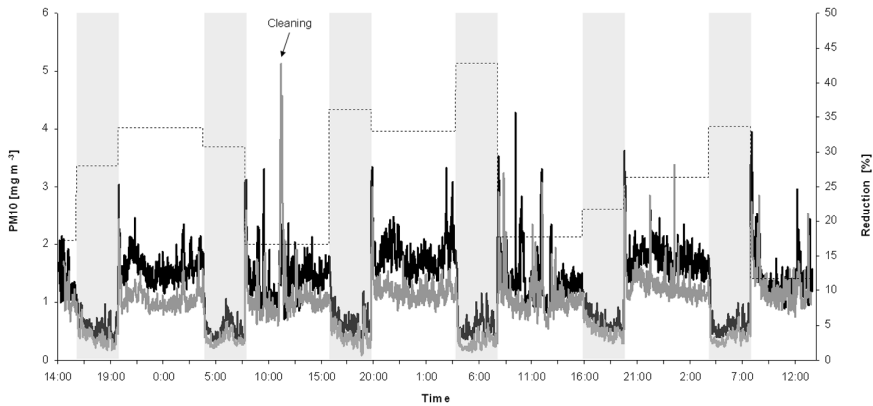
Figure 6.4. Average PM10 concentration pattern during 72 h, starting on day 31 of the first rearing cycle for control (black line) and ionization (grey line) rooms before and after cleaning ionization system. PM10 concentration reduction percentage is shown as dashed line. Dark periods are shown as shaded blocks. Note two dark periods, one from 03:45 to 07:45 a.m. and another from 15:45 to 19:45 p.m.

Particle size distributions were similar in control and ionization rooms. Particle counts per size range were on average 41% lower in ionization compared with control rooms. The relative difference in particle counts in control and ionization rooms in all measured size ranges decreased in time. On day 5, reduction in particle counts was the highest (76% for all size ranges), and it then decreased to 28% (day 19), 36% (day 26), and 24% (day 34) (Figure 6.5). At the start of the rearing cycle (days 5 and 26), a comparable level of reduction was observed between particles bigger and smaller than 10 μm. By the end of the rearing cycle (days 26 and 32), reduction in particles smaller than 10 μm decreased, being approximately two times lower compared with reduction of number of particles bigger than 10 μm.

Analyzed per size range, the statistical differences between average particle counts in control and ionization rooms also varied during the rearing period. On day 5, particle counts were higher ($P < 0.05$) in control than in ionization rooms for all size ranges, except for those larger than 12.5 μm. On day 19, particle counts were only significantly higher in the control room ($P < 0.05$) for size ranges between 0.8 to 1 μm, and between 7.5 to 8.5 μm. On day 24, particle counts were higher ($P < 0.05$) for size ranges from 7.5 to

30 μm, except for the size ranges from 8.5 to 10.0 and from 20 to 25 μm. Over the entire measured range (0.25 to >32 μm), two high reduction peaks can be observed in Figure 6.5, one for particles between 0.58 and 1 μm, and one for particles between 7.5 and 30 μm. The middle particle size ranges (from 1.30 to 2 μm), and the smallest ranges (0.28 to 0.35 μm) showed the lowest average reductions.
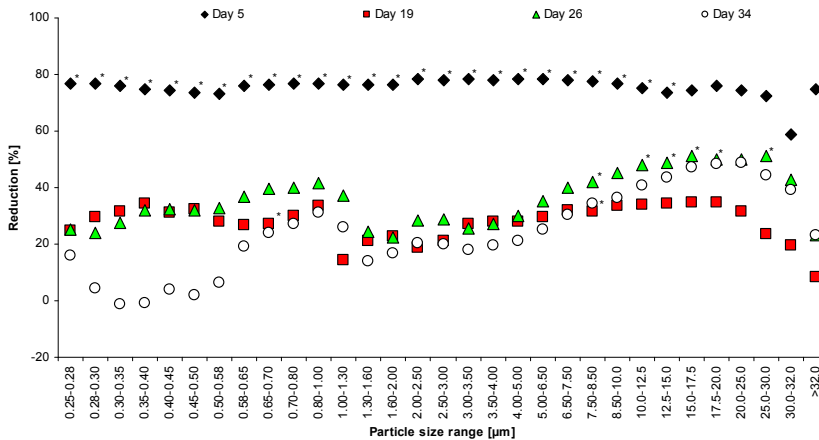

Figure 6.5. Average reduction (%) of particle counts, per size range from 0.25 to >32 μm, between the control and ionization rooms, on days 5, 19, 26 and 34, of the second rearing cycle (* indicates significant difference between control and ionization conditions at P < 0.05).

At human's breathing height, PM10 concentrations measured during personal PM10 sampling, were on average (SD) 29% (7%) lower in ionization rooms compared with control rooms, similar for both rearing cycles. Total average (SD) PM10 concentration for the whole experiment in ionization rooms was 1.7 (0.8) mg m^{-3}, compared with 2.4 (1.1) mg m^{-3} in control rooms. There was an increase in PM10 concentration also at human's breathing height along time, followed by an increase in PM10 concentration reduction. Average (SD) PM10 concentration in ionization rooms was 1.2 (0.6) mg m^{-3}, compared with 1.6 (0.7) mg m^{-3} in control rooms on day 25. On day 32 average (SD) PM10 concentration in ionization rooms was 2.3 (0.7) mg m^{-3} in ionization rooms, compared with 3.2 (0.9) mg m^{-3} in control rooms.

6.3.3. Airborne micro-organism emissions

We found no effect of ionization on micro-organism concentrations and emissions. Average (SD) total bacteria concentrations were 6.7 (0.4) log cfu m^{-3}, ionization; 7.1 (0.2) log cfu m^{-3}, control, during the first rearing cycle. Average (SD) total bacteria concentrations were 6.4 (0.3) log cfu m^{-3}, ionization; 6.5 (0.4) log cfu m^{-3}, control, during the second rearing cycle. Average (SD) total bacteria emissions were; 24.6 (6.5) log cfu h^{-1} $bird^{-1}$, ionization; 25.4 (9.5) log cfu h^{-1} $bird^{-1}$, control, during the first rearing cycle. Average (SD) total bacteria emissions were 20.8 (8.4) log cfu h^{-1} $bird^{-1}$, ionization; 20.2 (7.1) log cfu h^{-1} $bird^{-1}$, control, during the second rearing cycle. Average total bacteria emissions generally increased during the rearing period ($P < 0.001$).

Average (SD) airborne fungi and mold concentrations were 3.0 (0.6) log cfu m^{-3}, ionization; 2.9 (1.0) log cfu m^{-3}, control, during the first rearing cycle. Average (SD) airborne fungi and mold concentrations were 5.7 (0.5) log cfu m^{-3}, ionization; 5.8 (0.4) log cfu m^{-3}, control, during the second rearing cycle. Average (SD) airborne fungi and mold emissions were 11.5 (4.8) log cfu h^{-1} $bird^{-1}$, ionization; 10.9 (4.2) log cfu h^{-1} $bird^{-1}$, control, for the first rearing cycle. Average (SD) airborne fungi and mold emissions were higher for the second rearing cycle, 18.2 (7.2) log cfu h^{-1} $bird^{-1}$, ionization; 18.0 (6.4) log cfu h^{-1} $bird^{-1}$, control. Average airborne fungi and mold emissions also increased during the rearing period ($P < 0.05$).

6.3.4. Ammonia emissions

Total NH_3 emissions per bird per day were similar in ionization rooms (0.16 g $bird^{-1}$ day^{-1}) and control rooms (0.17 g $bird^{-1}$ day^{-1}) rooms, during the first rearing cycle. During the second rearing cycle, total NH_3 emissions were slightly lower than those measured during the first rearing cycle, but also similar in the ionization rooms (0.12 g $bird^{-1}$ day^{-1}), and control rooms (0.10 g $bird^{-1}$ day^{-1}). Dynamics of emissions were similar in ionization and control rooms during the rearing cycles, especially during the first 20 days in both rearing cycles (Figure 6.6). Daily variations were observed, with emissions following the same trend as ventilation exchange rates, increasing during light periods and decreasing during dark periods in all rooms. After day 20, some rooms showed different NH_3 emissions. Emissions decreased in

ionization and control rooms after day 20 in the second rearing cycle, compared with the first rearing cycle. These differences are attributable to differences in ventilation exchange rate. Cumulative NH_3 emissions during the rearing period were higher in the first rearing cycle (15 kg) compared with the second rearing cycle (10 kg).
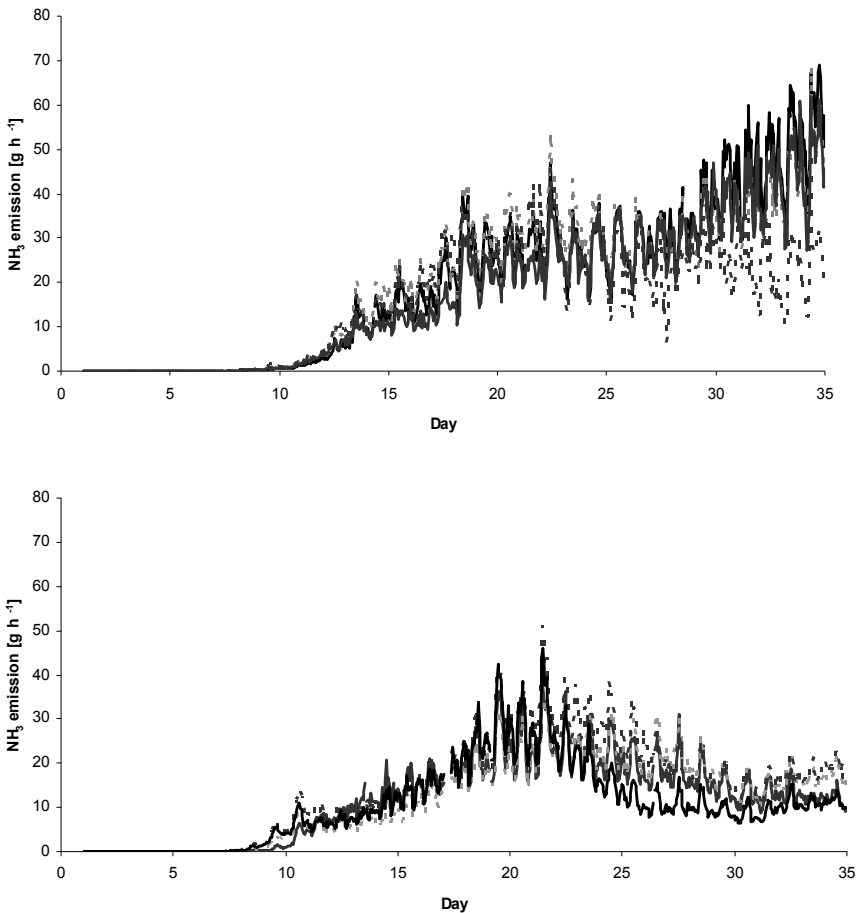

Figure 6.6. Average daily ammonia emission rate (g h^{-1}) along the rearing cycle for the first rearing cycle (top), and for the second rearing cycle (bottom). Dark continuous lines show control and dashed grey lines show ionization rooms.

6.3.5. Odor emissions

We found no effect of ionization on odor emission. Average (SD) odor emissions were 1.0 (1.6) ou$_E$ s^{-1} bird^{-1}, ionization; 0.9 (1.8) ou$_E$ s^{-1} bird^{-1},

control, for the first rearing cycle. Average (SD) odor emissions were 1.2 (1.7) ou_E s^{-1} $bird^{-1}$, ionization; 1.1 (1.6) ou_E s^{-1} $bird^{-1}$, control, for the second rearing cycle.

6.3.6. Performance of ionization system

The ionization system used in this experiment worked properly over the whole experiment in terms of ion concentrations. Ion concentrations for both ionization rooms were approximately 1800 ions cm^{-3}, ranging from 220 to 6400 ions cm^{-3}. Ion concentrations did not vary in time, and remained more or less constant along the whole experiment. In each room, however, ion concentrations were not uniformly distributed, showing maximum ion concentrations closer to discharge electrodes.

Ozone concentrations in all rooms were below 0.01 ppm in both rearing cycles. Ozone concentrations remained below the lowest detection limit of the ozone tubes. No differences were found between ionization and control rooms. Over the first days of the rearing cycle, however, ozone could be detected in ionization rooms, perceived as an intense smell of "clean bed sheets or fresh forest air". As ventilation rates increased along the rearing cycle, this smell could not be perceived after day 5.

Ultra fine particle counts were on average (SD) 45% (19%) lower in ionization compared with control rooms. Total average (SD) ultra fine particle counts for the whole experiment in ionization rooms were 8563 (9654) particles cm^{-3}, compared with 15,641 (16,618) particles cm^{-3} in control rooms. Concentration of ultra fine particles decreased during the rearing period. Average (SD) ultra fine particle counts were 6308 (2553) particles cm^{-3} ionization; 16,569 (7113) particles cm^{-3}, control, during the first rearing cycle. Average (SD) ultra fine particle counts were 9216 (10,797) particles cm^{-3} ionization; 15,394 (18,341) particles cm^{-3}, control, during the second rearing cycle.

It was observed that dust layer increased during the rearing period, and reached approximately 1 cm thickness on some surfaces (e.g. grounded metal surfaces such as the feed silo or feed storage bin) towards the end of the rearing cycle. Dust deposited on room surfaces was evident after the first week of the rearing cycle. Dust was attracted to the collection plates as

well as other grounded surfaces. These surfaces were generally plastic or metallic. Visually, the difference between ionization and control rooms became more evident during the rearing period, as concentrations of dust in the rooms increased. Ionization rooms showed a light yellow color because of dust deposition on walls and especially dust deposition on the protection cover of the tube light armature.

The increase of accumulated dust on room surfaces coincided with a decrease in current of the system. During the first rearing cycle, amperage showed a linear decrease in time. Amperage was set to 0.7 mA at the start, but it gradually decreased during the rearing cycle, showing a minimum of 0.4 mA on the last days (days 31 and 32). During the second rearing cycle amperage was stable at a level of 0.9 mA. Cleaning of the grounded plates showed a very weak effect on amperage which increased slightly after cleaning. The amount of total dust collected and brushed off the aluminum plates at the end of the second rearing cycle was on average (SD) 2449 (70) g in ionization rooms.

6.3.7. Bird performance

No statistically significant differences in broiler performance data (weight gain, feed and water consumption, and mortality) were found between ionization and control rooms. Also no differences were found in foot pad lesions between ionization and control rooms. Total average (SD) weight gain for the whole experiment in ionization rooms was 57.1 (0.5) g day^{-1} bird^{-1}, compared with 57.0 (1.0) g day^{-1} bird^{-1} in control rooms. Average feed conversion (SD) was 1.639 (0.015) kg feed kg bird^{-1} in ionization rooms, compared with 1.633 (0.017) kg feed kg bird^{-1} in control rooms. Average water consumption (SD) was 161.8 (1.7) mL day^{-1} bird^{-1} in ionization rooms, compared with 161.4 (2.4) mL day^{-1} bird^{-1} in control rooms. Average mortality (SD) was 2.9% (0.3%) in ionization rooms, compared with 2.8% (0.7%) in control rooms. Average foot pad lesions score was 86 at broiler house and 106 at slaughterhouse in ionization rooms, compared with 88 at broiler house and 103 at slaughterhouse in control rooms.

6.4. Discussion

Overall measured mass reduction for PM emission in our experiment was 36% for PM10, and 10% for PM2.5. At a human's breathing level, reduction for measured PM10 concentration was approximately 30%. Reductions in PM concentrations indoors are in agreement with lower ranges of reductions reported for PM concentrations in other studies. Higher reductions of PM concentrations, 43% in a broiler house (Ritz *et al.*, 2006), and 61% in a broiler breeder house (Mitchell *et al.*, 2004) have been reported. These higher reduction percentages are probably only referred to PM10, as regards to different PM fractions (PM10 and PM2.5), as in our experiment. Particulate matter was furthermore measured in these studies with light scattering devices. Light scattering devices could be affected by particle charges, as they have plastic sampling inlets, usually positively charged, which could cause attraction of negatively charged particles, and thus loss of particle mass measurement in the tested houses (Lyngtveit and Eduard, 1997). When using gravimetric analysis to measure PM mass, however, this effect is less probable because the electrical charge is smaller (Lyngtveit and Eduard, 1997). Differences in ionization system lay-out in these studies compared with ours in terms of dimensioning and positioning of the system could also play a role.

There are a few data available on PM10 and PM2.5 emission reductions from poultry houses using ionization, although it is better to evaluate and compare PM reduction efficiency attributable to ionization systems based on emission rates, rather than based on concentrations, because concentrations can be affected by ventilation rates (Lim *et al.*, 2008). Reductions in PM10 emissions of 47% were observed in a laying hen house (Lim *et al.*, 2008). Higher reductions in PM10 emissions (from 71 to 75%) were reported in a dairy cow house (Dolejs *et al.*, 2006). These higher reported emission reductions compared with our experiment could be related to lower average PM10 concentrations (approximately ten times lower) in the dairy cow house, and lower ventilation exchange rates (1141 m^3 h^{-1}) compared with those registered in our experiment (4757 m^3 h^{-1}).

We found a clear difference in PM emissions between the first and the second rearing cycle, although overall, PM concentrations and emissions

were comparable with those reported in other studies in broilers (Lacey *et al.*, 2003; Roumeolitis and Van Heyst, 2007). The higher humidity levels and the lower dry matter content of the litter observed in the second rearing cycle might be the main cause of the lower emissions in the second rearing cycle. Also NH_3 emissions were noticeably lower in the second compared with the first rearing cycle. This is not in agreement with findings of Groot Koerkamp *et al.* (1996) where NH_3 emission decreases at higher dry matter content of the litter. It might be, however, that the wet upper layer of the litter formed a crust which prevented NH_3 being emitted from the bottom layers of the litter. A similar pattern was described by (Calvet, 2008) in broilers during winter NH_3 measurements, with low ventilation rates, and as a result, low dry matter content of the litter, with lower NH_3 emissions compared with summer measurements in the same house.

Reductions of PM10 emissions were more stable along the rearing cycle than PM2.5 reductions which decreased with age of the birds. In growing piglets a decrease in reduction of PM concentrations when using ionization was observed after the third week of the rearing period, and furthermore showed negative reductions by the end of it (weeks 6 and 7) (Tanaka and Zhang, 1996). Such results could be explained by three processes: higher ventilation exchange rates towards the end of the rearing period, hence, higher air velocities and a lower probability that charged particles are reaching grounded surfaces before being emitted; a decrease in free ion concentrations in the air in ionization rooms with increasing PM concentrations and ventilation rates; and/or increasing layer of deposited dust on room surfaces along time and decreasing attraction of dust to these surfaces. The two steps involved in particle removal being particle charging and electromigration of charged particles due to electric fields, as described by Mayya *et al.* (2004), could be considerably affected by these three processes: high ventilation rate, decreasing ion concentrations, and increasing dust layer.

A considerable effect of ventilation exchange rate and the thickness of the layer of dust, and a negative relation between these two situations and PM reductions has also been stated in other studies (Bundy, 1984; Nicolai and Hofer, 2008; Tanaka and Zhang, 1996). Particulate matter removal efficiencies decrease as air circulation rates increase because increased

ventilation generally reduces ion density and residence time (Mitchell, 1997; Tanaka and Zhang, 1996).

Also, the electrostatic voltage is related to the thickness of the deposited dust layer and the electrical resistance of dust, and it increases as more dust is accumulated on collection plates or grounded surfaces (St George and Feddes, 1995a). High electrostatic voltage difference between the building surfaces and dust layer can insulate the surfaces and reduce the attraction of airborne dust to building surfaces (Tanaka and Zhang, 1996). Results obtained by Bundy (1984) in chamber experiments show similar trends in decreasing electrical field strength between the deposited layer of particles and the ground, as the layer of dust increases on the collector plates. To counteract this effect, a higher voltage can be applied to the discharge electrodes, although increasing the risks of ozone formation (Boelter and Davidson, 1997; St George and Feddes, 1995b), and undesired charging of objects in the rooms because of high electrostatic electricity level (Grabarczyk, 2001).

In our experiment, minimal effect of cleaning of the grounded collection plates could be observed on PM10 concentrations. The use of mechanical cleaning and dust collection used in the second rearing cycle did not to improve the reduction efficiency overall compared with only brushing off. Results of an ionization system installed in a commercial hatching cabinet where grounded plates were automatically rinsed and cleaned every 10 min, showed three times higher reductions of PM compared with our experiment (Mitchell and Waltman, 2003). Hence, our results could presumably change when more area in the rooms was cleaned (e.g. ceiling and walls), although it was not considered a practical measure to be applied.

The effect of ionization on reduction of PM emissions was highest for particles in the upper size ranges (from 7.5 to >32 µm), compared with lower size ranges. Higher reduction efficiencies of ionization in relation to increased particle size have been reported in other studies. Reductions of total suspended particles in a pig house were 30% higher than reductions of PM10 and PM2.5, although no relevant differences were found between reductions of PM10 and PM2.5 (Nicolai and Hofer, 2008). Higher reductions in particles bigger than 3 µm (Rosentrater, 2003) and bigger than

5 μm (Tanaka and Zhang, 1996) compared with smaller particles, have also been reported when testing ionization systems in livestock houses.

Reduction of particles by ionization systems has been reported to be a size-dependent process (Grabarczyk, 2001; Mayya *et al.*, 2004). Furthermore, the average calculated PM2.5 reduction in our experiment at normal scale (10%) was considerably lower than that for PM10. The reason is there are distinct particle charging mechanisms acting on small particles (<0.1 μm) which are charged by thermal charging mechanisms; compared with bigger particles (> 0.5 μm) which are charged by field charging mechanisms. In thermal charging, the charge acquired by particles is proportional to the diameter, whereas in field charging, it is proportional to the square of the diameter (Bundy, 1984). Another possible explanation for the differential effect of ionization with regard to particle size is the higher probability of ions being attached to bigger particles than to smaller particles. The reason is the extent to which ions can attach to particles depends on particle size and shape (Kunkel, 1950), the mean particle size being generally bigger in PM10, compared with PM2.5. The higher number of particles in the lower size ranges compared with bigger size ranges could also affect particle charging, as given a constant ion concentration, a relatively smaller percentage of particles will be charged in those size ranges with higher numbers. A higher reduction in concentrations of big particles (> 2 μm), may also reduce the probability of aggregation of small particles to big particles and sedimentation, and so cause a decrease in reduction of the small particles (Tanaka and Zhang, 1996).

No significant effect of ionization on airborne micro-organism, NH_3, or odor emissions was observed. Control levels of airborne micro-organisms and NH_3 were in the ranges presented in other studies (Lacey *et al.*, 2003; Seedorf *et al.*, 1998; Wheeler *et al.*, 2006). For odor, control levels were above the reference level of 0.24 ou_E s^{-1} $bird^{-1}$ for Dutch broiler houses (Infomil, 2009). Some studies have presented reductions of NH_3 when using ionization in broiler houses (Mitchell *et al.*, 2004; Ritz *et al.*, 2006). However, results are ambiguous, with reductions ranging from 13 to 56%. Reductions in odors and NH_3 could be expected, when a high proportion of these compounds found in the air of livestock houses would be adsorbed on PM. Despite research having shown that this proportion can be high (Koziel *et*

al., 2007; Takai *et al.*, 2002), the main part of NH_3 and likely odors are found in gaseous form. Nevertheless, olfactometry techniques used in our experiment require dust filtering, thus they do not include the effect of dust on odors. Although the reduction in PM concentrations in ionized rooms is the most probable mechanism for reducing odors in such rooms, our results as regards to the effect of ionization on odors, did not consider the odor included in PM. Overall, our results did not show any effects of PM removal on these pollutants.

Although ions have furthermore been reported to have a potential to kill micro-organisms and reductions in microbial load using ionization have been observed in some studies (Chiumenti and Guercini, 1990; Grinshpun *et al.*, 2004; Holt *et al.*, 1999), we did not find any significant differences in micro-organism emissions. Reduction of airborne micro-organisms in livestock houses using ionization have been reported in the range from 33 to 96%, normally exceeding PM reduction efficiencies. The reason for this discrepancy could be because of the sampling system used in our experiment, because impingement into liquid media tends to give higher colony counts for environments where micro-organisms are carried as aggregates, compared with impaction on culture plates, as used in most of the other studies.

The ionization system tested in this experiment did not produce high levels of ozone, nor did it increase ultra fine particle formation, despite these are common side effects reported in literature. Ozone concentrations could not be measured below 0.01 ppm by detector tubes, although ozone was perceived as a smell at the start of the cycle. The detection limit for human's nose is in the range from 0.01 to 0.03 ppm. Other studies have reported similar ozone concentrations, usually in the range from 0.01 to 0.165 ppm, although this depends on the type of ionizer and room (Britigan *et al.*, 2006). Ultra fine particle concentrations were lower in ionization rooms compared with control rooms. Results suggest a low rate of ultra fine particle formation in ionization rooms, below the rate of ultra fine particle formation in control rooms. Ion levels were stable in time.

Bird performance was slightly above the ranges presented in other studies (Al Homidan *et al.*, 1998; Feddes *et al.*, 2002). No significant effect of

ionization on any of the tested bird performance variables nor on foot pad lesions were found, despite there is evidence that increased negative air ion levels can have some beneficial effects on animal performance, whereas depleted ion levels of both polarities, or increased positive ion levels, result in no change or in a reduction in performance. These effects have been tested in mice, showing changes in mortality rates (Krueger and Reed, 1976) and in chickens, showing higher growth rate in animals exposed to a negatively ionized environment (Quarantelli *et al.*, 2000).

To compare the performance and pollutant reduction efficiencies of ionization systems used in livestock houses is complicated because the number of factors affecting, as well as differences in lay-out, maintenance and cleaning of the system, and measuring techniques used in evaluation. To compare PM reduction efficiencies, a comparable expression is necessary. This expression could include variables such as generated PM per time unit and per ionization electrode. As a result, it is still difficult to predict performance of ionization systems and its effect on PM and other pollutants in commercial livestock houses.

6.5.   Conclusions

We evaluated the effect of air ionization in reducing atmospheric pollutant emissions in a pilot scale broiler house with four identical rooms during two consecutive rearing cycles. Two of these rooms were randomly assigned to the ionization treatment, while the other two rooms served as control. Furthermore we evaluated the performance of the tested ionizations system and its influence on bird performance. From our results, we can conclude that:

- Ionization system effectively reduced total PM10 mass emissions by 36% (SD 2%), and PM2.5 emissions by 10% (SD 33%).

- Particulate matter reduction was higher at the start of the rearing cycle. Reductions of PM10 emissions were more stable along the rearing cycle than PM2.5 reductions which decreased with age of the birds.

- Reductions might have been negatively affected by increasing ventilation rates, PM concentrations, and dust accumulation on room surfaces.

- Higher reduction efficiencies of ionization in relation to increased particle size were observed. Particles in the upper size ranges (from 7.5 to >32 µm) were more effectively reduced than smaller PM.

- Ionization did not have a significant effect on micro-organism, odor or NH_3 emissions reduction.

- Ionization did not show any significant effect on bird performance or on foot pad lesions.

- Ionization proved to be an effective and practical system to reduce PM emissions, with minimal maintenance and labor needs. There is need to evaluate the use of ionization in commercial broiler houses to validate these results, and the performance of ionization systems and its effect on PM and other pollutants, in real conditions, in bigger enclosed spaces.

6.6.   Acknowledgements

6.7.   References

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Chapter 7

General discussion and conclusions

7.1. General discussion

Livestock production systems are nowadays considered as important point sources of aerial pollutants because economies of scale have resulted in fewer farms of bigger size, that are geographically more concentrated in smaller areas (Aneja *et al.*, 2006; EPA, 2002). In certain European regions where background PM concentrations due to other sources are already high, such as The Netherlands, Flanders, North Italy, and North-East Spain amongst others, PM emitted from livestock houses can cause exceedance of the limits established by the European air quality regulations. In the last 5 to 10 years, there has been an enforcement of environmental regulations which challenge the livestock sector and the present production systems. Together with ammonia, particulate matter (PM) from livestock housing is a major concern due to its health and environmental adverse effects (Takai *et al.*, 1998). Therefore, PM is considered as one of the most important pollutants associated with livestock husbandry, with poultry and pig houses as the main contributors to PM emissions from livestock (EMEP-CORINAIR, 2007). Consequently, there is growing need to reduce atmospheric emissions from point sources such as intensive poultry and pig housing systems.

Modern livestock production systems have to produce profitable products, in the framework of food safety, animal welfare, and environmental protection. Therefore, the farming sector, local and national authorities are seeking for ways to achieve a more sustainable livestock production. The role of research in this field is to provide knowledge that can help in the decision-making to achieve this goal. Amongst these, abatement and prevention of environmental effects caused by livestock husbandry has until very recently been considered less important than food safety and animal welfare, perhaps because environmental pollution (especially atmospheric pollution) and its effects can go unnoticed and can not actually be seen.

As a consequence, livestock houses are still one of the most poorly characterized sources, and there is lack of knowledge which can allow making well-founded decisions to contribute to reduced livestock's emissions effectively. In the case of PM, this is aggravated because to date, we still do not know exactly the extent of the problem. It is not yet fully understood how PM is generated in livestock houses, what are the factors

influencing PM generation, and which sources to tackle. Furthermore, there is still a largely unknown and unquantified risk to the human population in the vicinity of livestock houses as regards to exposure of PM itself and to other pollutants bound to PM (gases and also micro-organisms) (Wathes *et al.*, 2004).

The aim of the research presented in this thesis was to acquire knowledge on where PM comes from in various livestock housing systems and to evaluate abatement techniques on reducing PM in relation with other pollutants. This study should contribute to improving the knowledge on main sources of PM in different housing systems, responsible for PM concentrations and emissions, and should supply necessary tools for improving actual reduction schemes. In this chapter, the main findings from each study described in this thesis are analyzed in a broader context and their implications for future research on how to control PM in and from livestock houses are given.

7.1.1. Source identification and quantification

Measures to control effectively PM from livestock production should include the following aspects: i) the quantitative estimation of the contribution of livestock production to PM concentrations in the ambient air; ii) the identification of the contribution of different livestock categories to PM in the ambient air; and iii) the knowledge of the contribution of each source within livestock houses to PM in the ambient air. These aspects are relevant to propose adequate abatement measures and can also be used to evaluate potential effects of PM. In this thesis we focused on source apportionment and on abatement of PM.

The knowledge on the contribution of each source within livestock houses to PM in ambient air involves the identification of the sources of PM emitted into the air. Sampling and analysis of particulate sources are required to apportion PM to the different sources. In this way, we analyzed sources that were representative of the material that reaches the ambient air in livestock houses. This was an essential starting point to carry out source apportionment of PM. In Chapter 3, the nature of morphological and chemical diversity in PM sources was analyzed.

Knowledge on particle morphology and chemical composition is also useful to understanding the potential health effects associated with exposure to PM from livestock houses. There is a great need to evaluate health impacts from exposure to PM emitted from livestock houses (Heederik *et al.*, 2007), but to date we still do not know what property of PM is responsible for toxic and deleterious effects. The toxic health effects of PM are likely to depend on several factors, including the size and composition of the particles, the level and duration of exposure, and age and sensitivity of the exposed person (Pope and Dockery, 2006). There are few studies that have documented the relationship between single chemical components of PM and health effects, but several studies have found that a the combination of various chemical components as well as physical properties of PM such as particle number, surface area, and mass, may affect toxicity of PM pollution (Harrison and Yin, 2000). Particles generated from combustion of fossil and biomass fuels, by high temperature industrial processes (such as smelting), products of chemical reactions in the atmosphere (such as SO_4^{-2} and NO_3^{-}), and fine particles from soil, are the established categories with chemical components likely to be responsible for negative health effects (Pope, 2000).

Although the mechanisms by which PM affects human health are not fully explained, consistent positive relationships have been established between inhaled fine (PM2.5) and coarse (PM10-2.5) PM and respiratory and cardiovascular disease, and mortality (Ballester *et al.*, 2002; Dockery *et al.*, 1993; Hoek *et al.*, 2000; Pope *et al.*, 2002). These studies, have dealt with mass exposure, expressing results on a mass basis (total amount than can penetrate into the lungs). However, expressed as particle number or surface area per unit mass, the amount of fine PM may be much larger than the amount of coarse PM (Brunekreef and Forsberg, 2005). Moreover, evidence of associations between ultrafine particles (particles less than 1 μm) and lung affections have been established, especially in areas with high traffic density (Seaton *et al.*, 1995). This can be explained by their small size and thus their ability to penetrate the lung wall and cause tissue damaging effects, and by their high surface area per unit of mass (Brown *et al.*, 2001). However, the number of ultrafine particles in the air and fine PM are poorly correlated and therefore ultrafine particles are unlikely to explain the relationship between fine PM and health effects on a mass basis. The focus of this thesis

was on primary coarse particles. In fact, most particles in livestock houses are coarse, and approximately only 15% of PM10 is PM2.5 (Cambra-López *et al.*, 2009); whereas in ambient urban air, this percentage can be higher than 65% (Harrison *et al.*, 1999).

As Harrison and Yin (2000) reported that the potential health hazard of PM is more related to particle number, surface area, and mass, rather than by its chemical composition, the results presented here open the door to improving the understanding and justifying the potential hazards associated with PM. Particle size and morphology, which is related with particle's aerodynamic behavior is very closely related to lung deposition. Furthermore, the four ways of particle deposition mechanisms in the human airways are: inertial impaction, sedimentation, interception and diffusion (Crowder *et al.*, 2002; Zhang, 2004). Particle's aerodynamic diameter determines the deposition mechanism. In general, when the aerodynamic diameter is larger than 1 μm, particles are deposited by the mechanisms of inertial impaction and sedimentation. Inertial impaction increases with particle velocity and mass. Sedimentation increases with particle size, particle density, and is a function of residence time in the airway (increasing when air velocity of breathing rate is low). Interception mechanisms occur with elongated particles (e.g., fibers), when the edge of the particle contacts the airway wall, and it is a factor of particle diameter. When aerodynamic diameter is smaller than 0.1-0.2 μm, diffusion is more important than the other mechanisms, and it increases as particle size decreases. The results presented here could be used in further studies to evaluate the relationship between these deposition mechanisms in the respiratory airways and different particle types which can be used to establish a closer relationship between the described particle morphologies and sizes in livestock houses and particle dynamics, in future studies.

In Chapter 5 we provided quantitative estimations of the contribution of different sources to airborne PM based on the knowledge of the most appropriate characteristics to use in source apportionment of PM in livestock houses identified in Chapter 4. We demonstrated that most particles come from dried manure, although the contribution of this source varied within livestock housing systems. In poultry houses, feathers and wood shavings were the second most important sources, while animal skin

was the second most important source in pig houses. Wood shavings in poultry and skin in pigs were especially relevant sources expressed in mass (because their big size). Sampling equipment used to measure PM is generally based on gravimetric methods which result in levels of PM expressed in mass as mg of PM per m^3 of air. When using sampling equipment based on methods other than gravimetry, variability of particle morphologies can explain why levels of PM measured with light scattering or tapered oscillating element microbalance (TEOM) are not comparable to gravimetric methods.

Results on quantitative estimations of source contributions suggest that the exposure to PM from livestock houses could aggravate health effects because manure particles are quite small. In pig houses, this effect could be even more important than in poultry, because the portion of manure falling into the PM2.5 fraction was shown to be higher than in PM10-2.5. Exposure to manure components implies an extra risk related to micro-organisms and fragments of these which are present in fecal particles, including mainly endotoxins (lipopolysaccharides) and beta-glucans (Donham *et al.*, 1986; Schulze *et al.*, 2006). These components are associated with lung infections and lung symptoms (Donham, 2010). Furthermore, exposure to poultry feathers, dander, and serum has been associated with allergenic reactions because inhalable poultry PM has been shown to contain several allergenic components (such as lipopolysaccharides found on Gram-negative bacteria, lipoteichoic acid derived from Gram-positive bacteria, and beta-glucans) which can induce antibody-mediated reactions in humans and birds (Bar-Sela *et al.*, 1984; Lai *et al.*, 2009). Although wood shaving particles only moderately contribute to PM in livestock houses, they also can be relevant to health effects, because softwood dust used as bedding (mainly from coniferous in poultry houses) is associated with skin disorders, rhinitis, and occupational asthma (Currie and Ayres, 2005; Douwes *et al.*, 2001). Our results suggest the risk and exposure to manure components in PM can be very high in and around livestock houses. Consequently, to reduce more effectively PM emissions from livestock houses, we should focus on manure because this would imply a double benefit: reducing contribution to ambient air PM pollution levels and reducing associated negative health effects of manure particles. In summary, the pathways by which PM can cause health

problems must be examined and the causal agents must be identified. One of the main gaps of knowledge is related to what are the main causal agents involved in health effects: is it the exposure to high levels of mass or number of PM? or is it the chemical composition? or is the ratio particle to surface area?

Furthermore, besides human health affections caused by PM in ambient air, animal health and welfare can also be affected by high levels of PM inside livestock houses and this may affect livestock performance and production efficiency (Curtis, 1972). The relationship between livestock performance and production efficiency and air quality has been reported in several studies (Al Homidan *et al.*, 1998; Al Homidan and Robertson, 2003; Donham, 1991; Feddes *et al.*, 1997). Donham (1991) reported positive correlation between PM concentration and disease in fattening pigs, as well as with slow weight gain rate. Guarino *et al.* (1999) identified a close link between increased PM concentrations and mortality in laying hens. Lai *et al.* (2009) stated that certain antigen components of PM may lead to an enhanced immune reactivity (hypersensitivity or allergies) and influence body weight in broilers. Therefore, air quality is not only a health and welfare issue, but also an economic issue and improving one could have added potential benefits on the other.

7.1.2. Control of PM at source

The results presented in this thesis don't make it easy to propose adequate abatement PM measures in livestock houses, because manure is an inevitable PM source and little can be done to avoid it. Nevertheless, in the light of these results, management practices which focus on improving housing cleanness and improving house design can have a potential to contribute to reducing PM levels. Other practices related to animal management and manure management can be helpful in this sense. Changes in environmental variables (i.e. reducing air temperature, increasing humidity, reducing wind speed, and improving ventilation rate) can have a potential to contribute to reduce PM levels. Control measures, however, must be compatible with livestock production and modifying environmental variables may be difficult because these factors also affect thermal comfort and economical performance of the animals (Banhazi *et al.*, 2008).

According to our results, there are three ways in which reduction of PM could be achieved: i) keeping manure humid, ii) reducing the time dried manure is in the rooms, and iii) reducing air movement and animal activity close to dry manure deposits. This makes reduction measures extremely site-specific, because the possibility of incorporating one of these management practices would depend on the type of housing. Moreover, although increasing manure humidity can lower PM levels, its side-effects on other pollutants such as ammonia should be considered, since it has been reported that ammonia volatilization increases with high levels of relative humidity in broiler litter (Weaver and Meijerhof, 1991). This contradiction in objectives could complicate reduction measures. For instance, manure drying systems in laying hens in battery cages has shown to reduce ammonia emissions by 6 to 27% compared with non-drying belt system (Groot Koerkamp *et al.*, 1995). As regards to PM, however, this would result in increased levels of PM (Kaliste *et al.*, 2004; Vucemilo *et al.*, 2008). Moreover, the suitability of animal welfare housing systems as regards PM is also questionable. For instance, changing from cages to bedded systems in laying hens, or from fully to partially slatted floor in fattening pigs, or from individual to group housing and straw bedding in sows can involve high PM levels, as it has been reported in several studies. Therefore, control measures should balance between all aspects. To be able to find the best balance, however, the inter-relationship between all air factors should be understood, and the combinations and synergies should be examined.

Nevertheless, keeping rooms clean is simple and can be achieved at low cost, with minimal labor. Banhazi *et al.* (2008) reported that increasing the cleaning regime and improving surface hygiene could have potential to improve air quality. Furthermore, our results suggest air cleaning techniques such as ionization can also achieve significant reduction levels at minimal costs. Although we did not found a significant effect of air ionization on bird performance, air ionization has also been reported to have beneficial effect on animal performance (Quarantelli *et al.*, 2000). This effect, however, is dependent of management conditions and can be higher under poor management conditions.

Animal's themselves can also contribute to PM levels, and the contribution of feathers (in poultry) and animal skin (in pigs) was shown to be relatively

abundant, especially in terms of mass contribution. Therefore, measures which focus on animals, such as mineral oil spraying, can be very effective. In pig houses spraying mixture of mineral oil and water has shown to reduce total PM from 30 to 85% with low maintenance costs (Kim, 2006; Takai and Pedersen, 2000). In broiler houses, high reductions were found, as well. The optimal oil dose is depending on the balance between PM reduction and animal welfare. PM reduction is increasing at higher oil treatment doses; foot pad lesions, however, are also increasing at high oil doses (Aarnink *et al.*, 2008).

Our results indicate that measures which focus on other sources such as bedding and feed will probably not have as high potentials to reduce PM levels in livestock houses as measures which focus on manure. In any case, use of "low-dust" feed would be more recommendable to be implemented in pig houses as regards poultry houses. As a rule of thumb, measures that have an effect in the 4 to 5.5 micrometer size-ranged particles could contribute to reducing PM levels because this is the average size of manure particles. At least, it can be expected that this would have a potential effect on particle mass. To reduce smaller particles (to improve the reduction of particle numbers), measures that favor particle aggregation and coagulation (e.g. ionization, and to a certain extent mineral oil and water spraying) could also have a high potential in this sense (see Chapter 6).

From our results a list of techniques which could have high potential in reducing PM in poultry and pig houses in the different housing systems evaluated in this thesis is shown in Table 7.1. Although measures to reduce at source are preferred because of its effects on improved air quality inside, when reductions achieved with these measures are not enough, end-of-pipe techniques which clean exhaust air and prevent PM from being emitted such as air scrubbers, can be very effective (Aarnink, 2007; Ogink *et al.*, 2008; Ogink and; Zhao *et al.*, 2008).

Table 7.1. Summary of available PM reduction options which could be implemented in poultry and pig houses in the different housing systems.

Livestock species	Housing system	Preferred reduction option
Poultry	Broilers - bedding	Prevent drying of litter Reduce activity Air ionization Oil spraying
	Laying hens - floor	Prevent drying of litter Increase removal frequency of manure Reduce activity Oil spraying
	Laying hens - aviary	Prevent drying of litter Increase removal frequency of manure Reduce activity Oil spraying
	Turkeys - bedding	Prevent drying of litter Reduce activity Air ionization Oil spraying
Pigs	Piglets- slatted floor Growing-finishing pigs - partially slatted floor Dry and pregnant sows - group housing	Improve pen design Improve feeding distribution and use of Pelleted feed Prevent pen fouling Reduce activity Oil spraying Oil on animals

7.1.3. The need for an integrated approach to evaluate abatement measures

Overall, the findings presented in this thesis have contributed to providing tools for designing better and more efficient PM reduction measures at source and for predicting how different techniques will work. However, evaluation on-farm is a critical step, and although in theory some measures can have a high potential, they should always be tested in practical conditions. In this case, our results suggest an integrated and whole-process approach, as carried out in Chapter 6, is necessary to draw conclusions.

Because PM is strongly related to other pollutants (i.e. ammonia, odorants and micro-organisms) measures that can reduce PM can also affect them. For this reason, reduction techniques should be evaluated in terms of their effect on PM levels, but also accounting for all aspects and relationships of PM. This will enable to understand better the relationship between PM and these pollutants which could represent a potential added value of a PM reduction option. Chapter 6 is a case-study where an integrated approach to evaluate a PM reduction option was used. Only in this way, can

recommendations be given whether or not to perform the next step: testing the option on real-scale farms. There is need to evaluate the use of reduction techniques in commercial houses to validate the results, obtained under experimental conditions, under real conditions. Only when assessing all aspects thoroughly and objectively, conclusions can be drawn with respect to the applicability of the reduction option at commercial farms.

An integrated approach should include measurements and evaluation of:

- Size-segregated PM concentrations (in numbers and in mass)

- Size-segregated PM emissions

- Effect on ammonia and other gases

- Effect on odors

- Effect on airborne micro-organisms (pathogenic and non-pathogenic)

- Livestock performance and productivity

- Economic evaluation

7.1.4. Directions for future research (Recommendations)

In this thesis, the initial objectives were fulfilled with certain limitations. These limitations are discussed below which can serve for improving future studies on source apportionment of PM from livestock houses and PM reduction:

- Improve resolution for particles having dimensions in the range of 0.1 to 1 µm. In this thesis (Chapters 3 to 5) these particles which are mostly composed of elements with low atomic numbers (i.e. light elements such as C, H, and O) were not included in single-particle analysis using SEM because they showed a low grey scale contrast with the filter background. This could result in an underestimation by number and composition, of light elements particles in the range of 0.1 to 1 µm, such as condensable organics, ammonium nitrates and sulphates, organic and elemental carbon. However, these particles are mainly secondary particles and in livestock houses,

most particles are primary coarse particles (bigger than 1 µm) and formed by mechanical processes.

- Identify how many and which type of bio-aerosols are present in PM from livestock houses: microbiological content of PM related to inhaling pathogenic bacteria and non-pathogenic bacteria (which can also be harmful); and their relationship with PM, because these microbial particles may exist as solitary particles or attached to larger ones.

- Determine composition of PM in terms of bioactive compounds (endotoxins, beta-glucans, antibiotics, allergens, and dust mites) which can increase the potential health hazard of PM.

- Realize an integrated evaluation of ionization in pig houses, which can be promising to reduce PM, and has proven to be technically feasible and practical, with PM reductions varying from 20 to 60% (Rosentrater, 2003; Tanaka and Zhang, 1997).

- Investigate other reduction techniques based on manure management practices and improved housing designs, which consider auto-cleaning systems and more frequent farm-cleaning programs, through complete and integrated evaluation and cross-effect assessment with other pollutants.

7.2.   General conclusions

The following general conclusions can be drawn from this thesis:

- Livestock production systems can emit considerable amounts of PM which have to be controlled and reduced to protect the environment and the health and welfare of humans and animals, and to comply with current European legislation on air quality.

- Specific methodologies for source apportionment of PM in livestock houses and standardized measuring protocols to measure PM levels and characterize the morphology and composition of PM in different size fractions need to be developed. Comprehensive field studies need to be performed, as well.

- The sources that can contribute to PM are specific for livestock housing systems and livestock species. Housing systems and livestock species determine particle diversity and heterogeneity.

- The experimental dust generation process was successfully applied to develop comprehensive morphological, chemical, and size single-particle characterization and analysis of PM from feathers, feed, manure, hair, skin, wood shavings and outside source collected from different housing systems for poultry and pig animal species.

- The qualitative morphological description of potential sources of PM from the surveyed poultry and pig houses in different housing systems and the presented particle-size distributions reported in this thesis, are valuable to compare similarities and differences in particle types and will allow faster and more accurate qualitative and semi-quantitative estimations of source contributions in future studies.

- To apply source apportionment models in poultry and pig houses, it is necessary to obtain particle chemical characteristics and although presence of N, Na, Mg, Al, Si, P, S, Cl, K, and Ca are found in most analyzed particles belonging to different PM sources, their relative elemental concentrations can be used to assign to the correct source from 58 to 62% of individual particles in poultry, and from 64 to 73% in pigs, in fine and coarse PM.

- Morphological particle characteristics can make additional value to using only chemical characteristics in source apportioning in poultry and pig houses when sources show distinctive and well defined individual particle morphology or differ in size.

- On average 69% of particles belonging to a mixture of sources from poultry and pig houses can be correctly assigned to their source based on the combination of chemical and morphological characteristics in fine and coarse PM, and based on our results, it is the recommended approach to apportion all individual sources to PM in livestock houses.

- In the surveyed poultry houses, source contributions vary amongst poultry housing systems, but most particles originate from manure (ranging from 9 to 85% in fine and from 30 to 94% in coarse PM) and from feathers (ranging from 4 to 43% in fine and from 6 to 35% in coarse PM).

- In the surveyed pig houses, source contributions vary amongst pig housing systems, but most particles originate from manure (ranging from 70 to 98% in fine and from 41 to 94% in coarse PM).

- When expressed in mass, big particles from wood shavings and especially from animal skin gain relative importance compared with number of particles.

- Air ionization can effectively and significantly reduce total PM10 mass emission by 36% and PM2.5 mass emissions by 10% % in broiler production, but it has no effect on airborne micro-organisms, odor or ammonia emissions.

- Overall, the studies presented in this thesis have provided new knowledge for better and more efficient designing of PM reduction measures at source and for predicting how different techniques will work. These measures, however, should finally be evaluated on-farm. This farm evaluation should not be limited to PM, but should include effects on other aerial pollutants, production results and economics, as well.

7.3. Conclusiones generales

De esta tesis pueden extraerse las siguientes conclusiones generales:

- Los sistemas de producción ganaderos pueden emitir cantidades importantes de material particulado ("particulate matter", PM) que debe ser controlado y reducido para proteger el medio ambiente y la salud y bienestar de las personas y animales, además de para cumplir con la legislación Europea actual sobre calidad del aire.

- Es necesario desarrollar metodologías específicas para realizar el reparto de las contribuciones de PM en alojamientos ganaderos, así como protocolos estandarizados para medir los niveles de PM y caracterizar la morfología y composición en distintas fracciones de tamaño del PM. Son necesarios también estudios de campo completos.

- La diversidad y heterogeneidad de las partículas de las distintas fuentes de alojamientos ganaderos está determinada por las propias fuentes que proporcionan el PM, que son específicas del sistema de alojamiento y de la especie animal.

- El proceso de generación experimental de partículas desarrollado fue adecuado para realizar una caracterización morfológica, química y de tamaño detallada de las partículas y el análisis de fuentes tales como plumas, pienso, estiércol, pelo, piel, viruta de madera y entorno exterior en las granjas, en diferentes sistemas de alojamiento de aves y porcino.

- La descripción morfológica cualitativa de las fuentes potenciales de PM de los alojamientos avícolas y porcinos en diferentes sistemas de alojamiento y la distribución del tamaño de las partículas de cada fuente fue útil para establecer similitudes y diferencias entre los tipos de partículas, lo que permitirá realizar estimaciones cualitativas o semi-cuantitativas más precisas y rápidas de las fuentes que contribuyen al PM en alojamientos ganaderos en trabajos futuros.

- Para aplicar modelos de reparto de las contribuciones de PM en alojamientos ganaderos, es necesario obtener las características químicas de las partículas; y aunque se ha detectado presencia de N, Na, Mg, Al, Si, P, S, Cl, K, y Ca en la mayoría de las partículas analizadas

procedentes de fuentes distintas de material PM, la concentración relativa de estos elementos puede utilizarse para asignar correctamente a cada fuente, desde el 58 al 62% de las partículas individuales en aves, y desde 64 al 73% en porcino, en el PM fino y grueso.

- Las características morfológicas de las partículas pueden aportar un conocimiento adicional respecto a las características químicas en el reparto de las contribuciones de PM en alojamientos avícolas y porcinos, cuando las partículas en cada fuente tienen una morfología individual bien definida y distintiva o difieren en su tamaño.

- Utilizando la combinación de las características químicas y morfológicas, se puede asignar correctamente a cada una de sus fuentes una media de 69% de las partículas procedentes de una mezcla de fuentes en alojamientos avícolas y porcinos en el PM fino y grueso. Según resultados obtenidos, este es el enfoque recomendado para repartir el PM generado en alojamientos ganaderos por las distintas fuentes individuales.

- En los alojamientos avícolas muestreados, la mayoría de las partículas se originan a partir de las plumas (rango entre 4 a 43% PM fino y entre 6 a 35% en el PM grueso) y de la gallinaza (rango entre 9 a 85% PM fino y entre 30 a 94% en el PM grueso).

- En los alojamientos porcinos, la mayoría de las partículas se originan a partir del estiércol (rango entre 70 a 89% PM fino y entre 41 a 94% en el PM grueso).

- Las partículas de viruta de madera y de piel animal adquieren mayor importancia relativa cuando se expresan estas contribuciones en masa de partículas.

- La ionización del aire pudo reducir eficazmente y significativamente la emisión total en masa de PM10 en un 36% y la de PM2.5 en un 10% en la producción de broilers, pero no mostró ningún efecto en los microorganismos suspendidos, sobre los olores o sobre le emisión de amoniaco.

- En su conjunto, los resultados presentados en esta tesis contribuyen a proporcionar unas herramientas básicas que permitirán diseñar unas

medidas de reducción de PM en origen mejores y más eficientes y, paralelamente, a predecir su funcionamiento. No obstante, estas medidas deberían evaluarse en última instancia en granja, y esta evaluación debería incluir el efecto sobre otros contaminantes aéreos, y también resultados de producción y económicos.

7.4. References

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