# On the Reproduction of Female Wild Boar (*Sus scrofa*) in Sweden

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Cover: Wild boar sow with piglets
(photo: A. Malmsten)

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## Abstract

Free-ranging wild boar (*Sus scrofa*) have been part of the Swedish fauna for thousands of years. Due to high hunting pressure and domestication, the species became extinct in the 17th century. After escapes from enclosures during the 1970-1980s, the numbers of wild boar in the southern part of the country increased steadily and today likely exceed 200,000 animals. To understand the population dynamics of wild boar, understanding of its reproductive period, oestrous cycle and reproductive potential is essential. The aim of this work was to increase the knowledge on female wild boar reproduction in Sweden.

The study was based on macroscopic examinations of reproductive organs (ovarian structures in relation to the appearance of, and findings in, the uterus) from 617 hunter-harvested female wild boar (>30 kg body weight), sampled in 2012 to 2015 on seven private estates, located in four Swedish regions. The age of the animals was determined, and body weight noted. The crown-rump length (CRL) of the embryos/foetuses was used to calculate the oestrous month and timing of expected farrowing. Sera were analysed by commercial enzyme-linked immunosorbent assays (ELISA) in order to investigate the prevalence of nine selected pathogens.

Uterus weight and length mirrored the present reproductive stage. In approximately 10 % of the examined animals, reproductive abnormalities such as return to oestrus, disturbed oestrous cycle, endometritis and embryo/foetal mortality were found. Of animals aged < 12 months, 11.4 % were classified as post-pubertal which was low compared to previous European studies. The proportion of post-pubertal animals increased with age and weight, and from summer to spring, suggesting a seasonal reproductive pattern. Although, the majority of all studied animals showed a seasonal reproductive pattern, farrowing also occured 'off-season'. The reproductive potential was high compared to other countries, and increased with age and weight, as did pregnancy rate. High sero-prevalence of Porcine parvo virus, Porcine circo virus type 2, *Erysipelotrix rhusiopathiae*, and *Mycoplasma hyopneumoniae* was found. No antibodies were detected against Porcine respiratory and reproductive syndrome virus, *Brucella suis,* or *Mycobacterium bovis.* In total, this work contributes to improved knowledge about wild boar reproduction in Sweden, which is of importance for wild boar management.



*Keywords:* oestrous stage, puberty, reproductive potential seasonality, serology.



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# Dedication

To Jonas, Vidar, Silje and Liv

Nothing Else Matters.

# Contents

# List of publications

This thesis is based on the work contained in the following Papers, referred to by Roman numerals in the text:

I Malmsten, A., Jansson, G. & Dalin, A.-M. (2016). Postmortem examination of the reproductive organs of female wild boars (*Sus scrofa*) in Sweden. *Reproduction in Domestic Animals*. Doi:10.1111/rda.12947.

II Malmsten, A. & Dalin, A.-M. (2015). Puberty in female wild boar (*Sus scrofa*) in Sweden. *Acta Veterinaria Scandinavica*, 58:55. Doi:10.1186/s13028-016-0236-1.

III Malmsten, A., Jansson, G., Lundeheim, N. & Dalin, A.-M. (2017). The reproductive pattern and potential of free ranging female wild boars (*Sus scrofa*) in Sweden. *Acta Veterinaria Scandinavica*, 59:52. Doi:10.1186/s13028-017-0321-0.

IV Malmsten, A., Magnusson, U., Ruiz-Fons, F. & Dalin A.-M. A serological survey of selected pathogens in wild boar (*Sus scrofa*) in Sweden. Submitted manuscript.

Papers I-III are reproduced with the kind permission of the publishers.

# Additional publication related to this thesis

- Malmsten, A., Dalin A.-M. & Pettersson, A. (2015). Caries, periodontal disease, supernumerary teeth and other dental disorders in Swedish wild boar (*Sus scrofa*). *Journal of Comparative Pathology*. 153:1, pp 50-56.

# Abbreviations

| | |
|---|---|
| ANOVA | Analysis of variance |
| *B. suis* | *Brucella suis* |
| bTB | *Mycobacterium bovis* |
| BW | Body weight |
| CL | Corpus luteum |
| CRL | Crown-rump length |
| *E. rhusiopathiae* | *Erysipelotrix rhusiopathiae* |
| ELISA | Enzyme-linked immunosorbent assay |
| FDW | Field dressed weight |
| IREC | Instituto de Investigación en Recursos Cinegéticos |
| *M. hyo.* | *Mycoplasma hyopneumoniae* |
| OR | Ovulation rate |
| PCV-2 | Porcine circovirus type 2 |
| PPV | Porcine parvovirus |
| PRRSV | Porcine respiratory and reproductive syndrome virus |
| SIV | Swine influenza virus |
| SVA | Swedish National Veterinary Institute |
| *T. gondii* | *Toxoplasma gondii* |

# 1 Introduction

## 1.1 Wild boar distribution

The wild boar (*Sus scrofa*), belonging to the family Suidae, is the ancestor to the domestic pig. It should not be confused with the 'feral hog' (or feral pig) that originates from domestic pigs that have escaped from farms. The wild boar has a widespread geographical distribution, inhabiting most areas in Europe, southern Asia, Northern Africa, and has also been introduced into North America (Figure 1).

Figure 1. Estimated wild boar distribution, native population (green), introduced population (blue, Wikipedia.org).

In many parts of Europe wild boar densities and distribution have grown considerably in the last decades, and the distribution of the species is difficult to confirm. In Sweden, free-ranging wild boar have been part of the fauna for thousands of years (see Emanuelsson and Liljegren, 2017). They have been hunted by humans for their meat, and other tissues have been used as tools (teeth, bones *et cetera*, Magnell, 2006). Even during the Stone Age in Scandinavia (4000 B.C.), wild boar were caught for domestication. Intensive hunting, as well as domestication, made the wild boar more or less extinct in Sweden in the 17$^{th}$ century (Markström and Nyman, 2002). The population that we see today in Sweden originates from an unknown number of animals that escaped enclosures during the 1970s and 1980s in the southern and middle parts of the country. The official management strategy following the escapes was aimed at culling all wild boar outside fences, but such efforts were unsuccessful. Instead, in 1987, the Swedish parliament decided that wild boar should once again be recognized as part of the Swedish fauna (Naturvårdsverket, 2010). Since the species' re-appearance, the population has increased, and today probably exceeds 200,000 individuals. Reliable estimates of population size are difficult to obtain at a national level. With a hunting bag of around 100,000 animals (Kjellsson, 2017) for several years in a row, the aforementioned estimate is not exaggerated. The current distribution of wild boar in Sweden ranges from Skåne in the south, to Gävleborg County in the middle part of Sweden (N55°-N61°, Kjellsson, 2017).

## 1.2 Wild boar biology

In areas where they are hunted, wild boar are nocturnal, which means that they are most active during the night. They are active in all seasons, with the activity range being influenced by minimum temperature, snow cover and humidity (Lemel et al., 2003). The species is an opportunistic omnivore, feeding on plants and animal matter. Invertebrates, mainly snails and earthworms, are more important than vertebrates (Massei et al., 1996; Herrero et al., 2006). They use their muscular, mobile snout and forefeet to root and scratch for feed. Wild boar have well developed sense of smell and hearing and rely more on these senses than on sight (Stegeman, 1938).

The social structure of the wild boar boar results in groups formed of several generations of females and their offspring (Dardaillon, 1988; Kaminski et al., 2005; Podgórski et al., 2014). Reproduction is synchronized within the female groups (Delcroix et al., 1990), which may play an important role in shaping this social structure. Adult males are mainly solitary and short-time social engagement is associated with mating, interactions with mating competitors, or enhancement of foraging (Podgórski et al., 2014).

## 1.3 Human aspects; Pros and cons of wild boar in Sweden

The perception of the general public in Sweden tends to be that wild boar are a nuisance, without regard to the animals' role as a resource. However, it is a popular game species, and high densities of wild boar are therefore desirable for many reasons, e.g. economic (hunting tourism), recreational, and access to game meat. During the hunting season 2012-2013, the total output of game meat in Sweden was 20,000 tonnes, of which 25 % was wild boar meat (Wiklund and Malmfors, 2014). Wild boar meat is considered an excellent and organic alternative to commercially produced meat. In addition, as with all non-ruminant game species, it has only a small impact on the climate (emissions of carbon dioxide, methane) compared to cattle farming. In recent decades in Sweden, several landowners have changed from mainly traditional farming to focus more on commercial hunting, i.e. offering hunts of free-ranging game including wild boar, fallow deer (*Dama dama*), and red deer (*Cervus elaphus*).

The presence of wild boar is also a source of conflict in the agricultural sector, as they may damage crops and pasture. The costs for wild boar damage to crops have been estimated to be 600-700 million SEK (€60-70 million) per year in recent years in Sweden [Pers. comm. Gren and Andersson, SLU, 2017]. In addition, traffic accidents involving wildlife have increased concomitantly with the number of wild boar. Between 2006 and 2016 the number of traffic accidents involving wild boar increased from approximately 1,020 to 4,757 (Nationella Viltolycksrådet, 2017). Moreover, the wild boar is considered as a potential reservoir of various pathogens that can infect other wildlife (Gortázar et al., 2007), domestic animals, and humans (see Meng et al., 2009 for review). Thus, dense wild boar populations may involve an increased risk of transmission of pathogens within the population, as well as from wild boar to other species.

## 1.4 Wild boar management and hunting legislation in Sweden

Regardless of whether wild boar are considered to be a nuisance or a resource, proper management plans ought to be devised and applied for wild boar populations. Today, wild boar in Sweden are managed by hunting and supplementary feeding.

In Sweden, wild boar hunting is permitted 24 hours a day, and use of hunting dogs is allowed between 1st August and 31st January. The hunting season of adult wild boar is between 16th April and 15th February. Hunting of yearlings is allowed year-round, but females with dependent piglets are always protected (Naturvårdsverket, 2017). Government and regional authorities have also made efforts to facilitate wild boar hunting by allowing the use of illumination at feeding sites, and night-vision equipment (Länsstyrelsen Kalmar län, 2017).

Although controversial, supplementary feeding is applied to a varying extent year-round throughout the wild boar range in Sweden. This makes studies of the direct effects of supplementary feeding on free ranging wild boar, including controls (i.e. areas without supplementary feeding), practically impossible to conduct in Sweden. The use of supplementary feeding during autumn and winter is mainly aimed at optimizing hunting and for hunting safety reasons. Feeding is also used to prevent starvation during the winter when other feed is limited. The motive during spring, summer and early autumn is to reduce wild boar damage to crops and pasture, i.e. to attract animals away from fields to feeding stations in the surrounding forest. The effect of such preventive measures is debated among hunters, farmers, policy makers, and researchers. While one European study indicated the success of supplementary feeding in reducing field damage locally (Andrzejewski and Jezierski, 1978), a more recent study did not reveal an effect (Geisser and Reyer, 2005). A Swedish study (Lemel, 1999) showed that the wild boars' attraction to supplementary feed seemed to vary with season and that they actually preferred maturing wheat and oats when available.

## 1.5 Female wild boar reproduction in Europe

Reproduction, besides migration and mortality, is a key component of population dynamics (Macchi et al., 2010). Thus, successful management of an animal species requires knowledge of its reproductive capacity.

### 1.5.1 Reproductive characteristics and reproductive failure

Even though the reproduction of female wild boar has been studied in a number of countries, e.g. France, (Mauget, 1972,1978,1982), Switzerland (Moretti, 1995), Spain and Portugal (Fernández-Llario and Mateos-Quesada, 1998; Fonseca et al. 2004), Czech Republic (Ježek et al. 2011) and Germany (Gethöffer et al. 2007; Frauendorf et al., 2016), few studies include information about the normal characteristics of their reproductive organs. It is not only important to obtain knowledge about the reproductive potential (e.g. variation, minimum, maximum) in a population, but also to gain knowledge about what could be expected (regarded as 'normal') before one can recognize any deviation from normal status levels. Abnormalities of reproductive organs may lead to reproductive failure and, if severe and frequent, the reproductive outcome of a population may be negatively affected. Knowledge about the occurrence and prevalence of reproductive failure should be of interest and importance for wildlife managers.

In domestic pigs, reproductive failure is the most common cause for culling sows and gilts in commercial herds (Engblom et al, 2007; Lucia et al. 2000; Stein et al; 1990). Reproductive failure can be manifested as absence of oestrous signs, regular or irregular returns to oestrus, conception failure, pseudo-pregnancy, abortion, mummifications, *et cetera* (Dalin et al. 1997; Engblom et al. 2007; Lucia et al. 2000; Stein et al. 1990). Congenital anomalies (Einarsson and Gustafsson, 1970) and cystic ovaries (Castagna et al. 2004; Dalin et al. 1997; Heinonen et al. 1998; Miller, 1984), as well as infectious diseases (see Pozzi and Alborali, 2012, for review), can all cause reproductive failure. The occurrence of reproductive abnormalities in the Swedish wild boar population is so far unknown, as is the prevalence of a number of pathogens that may affect reproduction directly or indirectly.

### 1.5.2 Reproductive seasonality

Based on European studies, pure breeds of wild boar without a history of hybridization with domestic pigs (which are considered to be poly-oestral throughout the year if not pregnant), have a seasonal reproduction pattern (Mauget, 1982, illustrated in Figure 2).

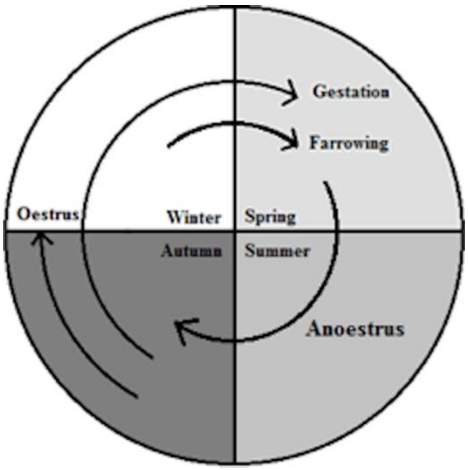


Figure 2. 'The wild boar year'. The seasonal order for the reproductive stages of female wild boar throughout the year, based on the results from Mauget (1982).

During the summer months, most female wild boar are anoestral (Mauget, 1982, Pépin et al., 1987, Delcroix et al., 1990). In southern and middle parts of Europe, the summer anoestrus coincides with high ambient temperatures, long days, and restricted access to feed (Mauget, 1978). In the autumn, as a result of shortened day length, the oestrous period begins (Mauget 1982). However, the timing of the first oestrus of the breeding season may vary between years, depending on the availability of natural feed and other environmental and climatic factors (Mauget, 1982, Delcroix et al., 1990, Macchi et al., 2010). For example, it appears that advancement or delay in the onset of breeding is related to the level of available feed.

Easy access to feed may cause a shortened anoestrous period (Pépin et al., 1987). Under normal conditions, the common pattern is one oestrous peak in the autumn/winter (Mauget, 1978). However, a bimodal pattern of oestrus and subsequent farrowing can also be seen (Mauget, 1972). In addition, some studies show that farrowing may occur throughout the year when feed is available all year round (Fonseca et al., 2004; Orlowska et al., 2012). In Sweden, hunter observations indicate that farrowing takes place all year around, but scientific data on the subject is missing.

### 1.5.3 Reproductive potential

The reproductive potential of wild boar, here defined and studied as the number of corpora lutea (CL) and the intra-uterine litter size, can be affected by many factors such as feed availability and climate (Servanty et al., 2007; Geisser and Reyer, 2005; Fernández-Llario and Carranza, 2000; Fernandez-Llario and Mateos-Quesada, 1998), as well as the genetic background and possible influence of domestic pigs in the wild boar population (Mauget, 1972). The weight and age of gilts and sows also affect litter size. Younger and lighter individuals normally produce smaller litters compared to older/heavier individuals (Fernández-Llario and Mateos-Quesada, 1998; Mauget, 1982). Studies have shown a correlation between supplementary feeding and litter size, with larger litter sizes in areas where supplementary feeding occurs compared to similar areas without feeding (Spain; Fernández-Llario and Carranza, 2000; Fernández-Llario and Mateos-Quesada, 1998).

### 1.5.4 Puberty in wild boar gilts

European studies have also shown a strong link between puberty and body weight, where wild boar in areas with sufficient access to feed attain higher body weight, and reach puberty at a younger age than animals in areas with less access to feed (Fernández-Llario and Mateos-Quesada, 1998; Massei et al., 1996). In female domestic pigs the definition of puberty is that the animal should show external signs of oestrus and ovulate (Evans and O'Doherty, 2001). Once puberty has been reached, the female should exhibit regular oestrous cycles if not pregnant. In Swedish domestic crossbreed pigs, gilts usually reach puberty at six to seven months of age (Dalin and Eliasson, 1987). However, in free-ranging wild boar it is difficult to monitor if a gilt shows oestrus and exhibits regular oestrous cycles. Therefore, most studies on puberty in free-ranging female wild boar are based on post-mortem macroscopic examination of the reproductive organs. Even so, the definition of puberty among these studies varies (Gaillard and Jullien, 1993; Ahmad et al., 1995, Gethöffer at al., 2007; Cellina, 2008), making comparisons difficult. The exact timing of when a female reaches puberty cannot be detected by macroscopic examination of reproductive organs. This method can only provide data on whether the animal has ovulated or not. If the exact timing of puberty needs to be studied, other methods such as repeated faecal (Hultén et al., 1995) or blood (Delcroix et al., 1990: Tast et al., 2000) sampling for progesterone analyses of fenced animals can be used. These methods are difficult or impossible to apply for free-ranging animals.

Knowledge of the reproductive seasonality, reproductive potential, and age at puberty is of importance for the management of the Swedish wild boar population. Knowledge of these parameters in combination with the sex and age composition in the population is necessary when modelling population growth. Moreover, knowledge of the reproductive potential is essential when planning culling strategies to meet specific management goals, such as increasing or decreasing the population density.

## 1.6 Knowledge of wild boar in Sweden

Previous Swedish studies regarding wild boar mainly describe wild boar movement patterns (Thurfjell et al., 2014), behaviour and habitat selection (Welander 2000; Lemel et al 2003; Truvé and Lemel, 2003; Thurfjell et al., 2009), and population dynamics (Lemel 1999). These studies include only limited information on reproduction, reproductive success, and factors affecting the reproductive potential of the species. Nonetheless, there are many diverging speculations and opinions (especially among hunters and farmers) on wild boar management in general, and reproduction in particular. These issues need to be addressed and resolved.

# 2 Aims of the thesis

The overall objective of this thesis was to provide knowledge on basic reproductive parameters of female wild boar in Sweden, based on macroscopic examination of the reproductive organs and sero-analyses, including:

- characteristics of reproductive organs in different oestrous stages
- abnormalities in the reproductive organs
- puberty in gilts
- reproductive seasonality
- reproductive potential
- sero-prevalence of reproductive pathogens

# 3 Materials and methods, including methodological concerns

This section provides a summary and comments on the materials and methods used in Paper I-IV. Detailed descriptions of the procedures performed are presented in the individual papers.

## 3.1 Study area and hunting methods (Paper I-IV)

The material and data for this thesis were collected between November 2011 and December 2015. Sampling was conducted during regular hunting on seven private estates, located in different regions (Skåne, Blekinge, Södermanland, and Uppland) in southern and central Sweden (Figure 3). Information on wildlife feeding practices, and estimated population sizes was provided by the wildlife manager on each estate. The size of the estates varied between 10 and 87 $km^2$ (non-fenced); the density of wild boar was estimated at 5-40 per $km^2$ (based on annual hunting bags and population monitoring at fields and feeding stations). Similar management strategies for the wild boar (hunting and feeding) were applied on all estates. The types of feed used at feeding stations were corn, grain, mixtures of cereal and sugar beets, and silage (wheat and oats, peas, and clover). Due to the integrity of the estates, no detailed information on what type of, or how much supplementary feed the different estates used will be presented here. Meteorological data (monthly mean precipitation and temperature) were obtained from local meteorological services (SMHI.se. 2016). During the study period, the weather conditions in the sampling regions were similar for all years with mean temperatures in January and July of -1ºC, and 18 ºC, respectively. The mean precipitation in January and July was 49 mm and 58 mm, respectively.

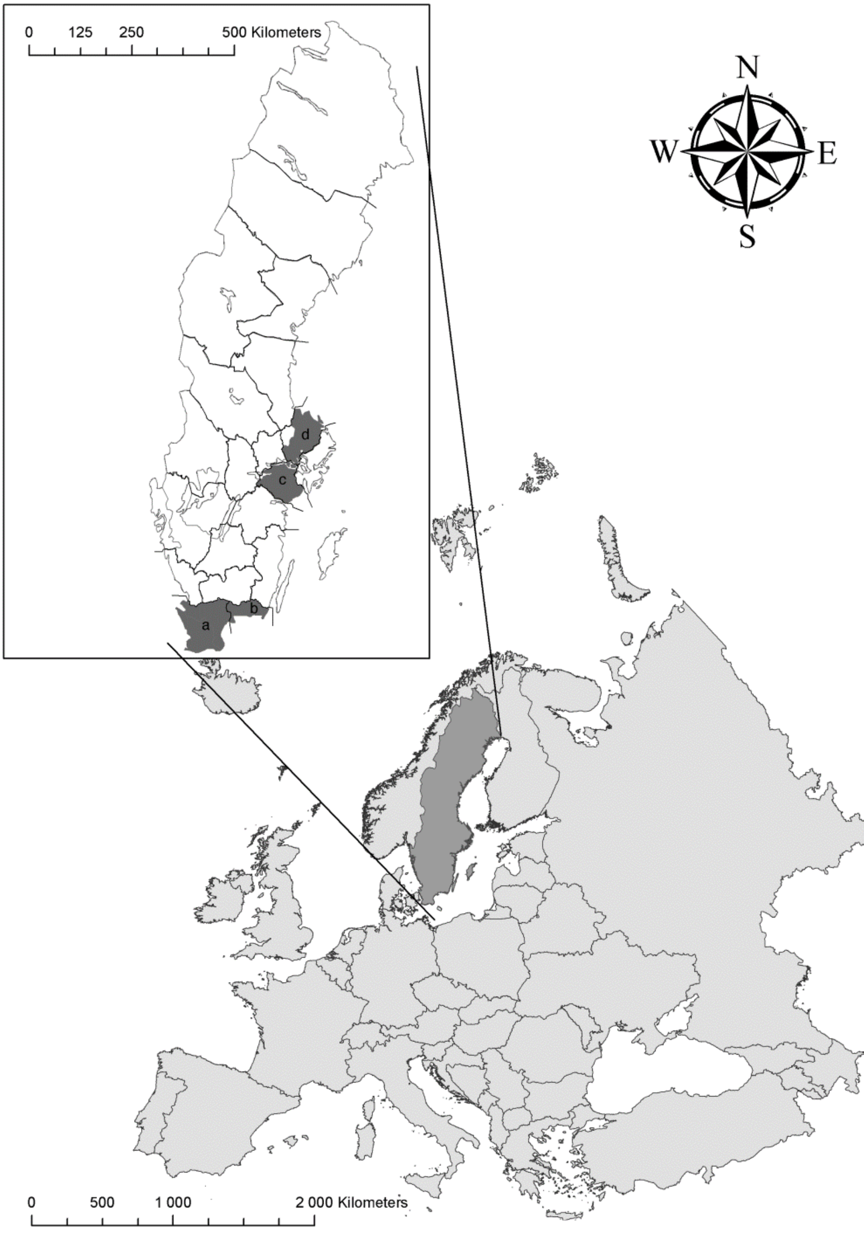




Figure 3. Map of Sweden highlighting the regions where female wild boar were sampled.
a) Skåne, b) Blekinge c) Södermanland, d) Uppland.

The hunting methods used were driven hunts and stalking. Driven hunts were the most common method in the study areas during the main hunting season (October until February) and could result in considerable daily hunting bags (30-100 animals). Stalking was used during spring and summer, and animals were most often shot while feeding in open field/grazing-ground (i.e. hunting mainly as a crop damage prevention measure).

## 3.2 Data collection (Paper I-IV)

The animals were sampled at the abattoirs of the estates within approximately two to four hours after culling. Date of harvest was noted. Body weight (BW) and field dressed weight (FDW, weight of the eviscerated animal with skin) were recorded. Initially, all females were sampled without selection based on BW. However, after examining 15 animals with a BW < 30 kg (range 14-29 kg) no ovarian activity was found, despite females with a BW <30 kg have previously been found to have reached puberty (Cellina, 2008). Based on the initial examination, we concluded that the probability of finding animals that had reached puberty in the lower weight category was very low to negligible. Therefore, a threshold was set and only females with BW above 30 kg were sampled. The age of each individual was estimated using tooth eruption and tooth replacement patterns, according to Matschke (1967).

The tissues collected for this project were the entire reproductive organs, blood, and stomach contents. Part of one ear, the spleen, and a kidney were also collected for different future analyses. The tissue and blood (= serum) samples were stored at -20°C until examination.

## 3.3 Sample criteria and sample size in the four papers

In total, 617 female wild boar were sampled. For Paper I, the criteria for the samples to be included were a complete set of reproductive organs with size and weight of ovaries and uterus, which were recorded for 569 of the 617 sampled animals. In Paper II, animals culled before January 2013 were excluded because they were not age-determined, resulting in 175 individuals that met the inclusion criteria, i.e. with a complete set of reproductive organs, age span 5 to 15 months, and weight information. For Paper III, individuals with a complete set of reproductive organs (575/617) were included in the study. In Paper IV, animals from estates from which we had the largest numbers of good quality (non-haemolysed) serum samples were included. In total, sera from 286 animals with known reproductive stage from three estates (in Blekinge, Södermanland and Uppland), were included for testing for the prevalence of antibodies against selected pathogens.

## 3.4 Macroscopic examinations of reproductive organs (Paper I-III)

The reproductive organs were examined macroscopically after thawing, according to Figure 4. The vestibule, vagina and cervix were cut open, and the colour of the mucosa and the presence of a hymen were recorded. The uterus (cervix excluded) was weighed, and the length of the uterus (one uterine horn and the corpus) was measured. The uterus was opened and the appearance of the endometrium [“colour” (haemorrhage graded as slight, moderate or high) and thickness (folding, presence of oedema)] was examined. Signs of pregnancy (embryos/foetuses) or signs of a previous pregnancy, such as thickening of the blood vessels at the connection between the uterus and mesometric ligament (Malmsten et al. 2014, Figures 5 and 6), thickness of the mesometric ligament, and width of the cervix, were recorded. Presence of ejaculate (gelatinous content from the male’s bulbo-urethral gland) in the cervix or uterus indicated that these animals were very recently mated (Figure 7).

The number and diameter of follicles (> 2 mm) and luteal structures in the ovaries were noted. Luteal structures were divided into fresh CL, regressed cyclic CL and regressed CL of pregnancy. The stages of the reproductive cycle were defined according to ovarian structures combined with the appearance and findings of the uterus. The classifications of the reproductive stages are provided in Table 1.

*Table 1. The classifications of the reproductive stages in female wild boar, according to ovarian structures, applied in this study (Paper I-III).*

| Reproductive stage | Structures in ovaries |
|---|---|
| Pre-pubertal | No CL, multiple follicles of small sizes |
| Prooestrus | Follicles ≥7 mm<br>(In animal with cyclic ovarian activity: presence of regressing cyclic CL about 6- 8 mm, yellow-white) |
| Oestrus | Mature follicles, about 9 - 10mm<br>(In animal with cyclic ovarian activity: presence of regressed cyclic CL about 5 mm, yellow-white) |
| Metoestrus | Fresh CL <5mm, small spots or with small blood filled cavity<br>(In animal with cyclic ovarian activity: presence of regressed cyclic CL 3- 4 mm, yellow-white) |
| Dioestrus | Early dioestrus, fresh CL 5 – 8 mm, in color pink with a blue red tone<br>Mid dioestrus, fresh CL about 10 mm, pink colour<br>Late dioestrus, fresh CL about 8 – 10 mm, pink-yellow colour |
| Pregnant | Fresh CL, about 10 mm, pink in color and some follicle of different size |
| Anoestrus | Few, small follicles<br>If presence of regressed CL of pregnancy, these structures have a dark color in the center of the cut surface, size vary depending on time from pregnancy, < 5 mm |

## Examination protocol

**Female wild boars**

ID nr:________________ Culling date:______________ Age:_________________________

Examiner:_____________ Date of examination:__________ Photo, motive:______________

___________________________________________________________________________

Organ examination

**Vagina**_________________________________________________________________

**Vestibulum**_____________________________________________________________

**Urinary bladder**_________________________________________________________

**Cervix**_________________________________________________________________

**Oviduct**________________________________________________________________

**Bursa ovarica**___________________________________________________________

**Left ovary**

Weight:__________g Length:_________mm Hight:_________mm Width:_________mm

___________________________________________________________________________

Follicles

Number:_________ Diameter largest follicle:__________mm Other:___________________

___________________________________________________________________________

Corpora lutea

Fresh: number__________ Diameter:__________mm Colour:________________________

Regressed: number__________ Diameter__________mm Colour______________________

Other:_____________________________________________________________________

**Right Ovary**

Weight:__________gr Length:_________mm Hight:_________mm Width:_________mm

___________________________________________________________________________

Follicles

Number:_________ Diameter largest follicle:__________mm Other:___________________

___________________________________________________________________________

Corpora lutea

Fresh: number__________ Diameter:__________mm Colour:________________________

Regressed: number__________ Diameter__________mm Colour______________________

Other:_____________________________________________________________________

**Uterus**

Weight:__________g Length left horn:__________cm Width:_________________________

Mucosa:______________________________________________________________________

_____________________________________________________________________________

Other:_______________________________________________________________________

**Embryo/Foetus** No left horn:_______________ No right horn_______________ st

| Allantochorin cm | Amnion cm | Fetal length cm | Fetal Weight g | Fetal sex f/m |
|---|---|---|---|---|
| | | | | |
| | | | | |
| | | | | |
| | | | | |
| | | | | |
| | | | | |
| | | | | |

_____________________________________________________________________________

_____________________________________________________________________________

**Oestrus stage**

| Prepubertal 0 | Anoestrus 1 | Prooestrus 2 | oestrus 3 | Metoestrus 4 | Dioestrus 5 | Disrupted cycle 6 | Pregnant 7 |
|---|---|---|---|---|---|---|---|
| | | | | | | | |

Comment:_____________________________________________________________________

_____________________________________________________________________________

Further investigation

Histology:__________________________________ Fix:______________________________

Virology:___________________________________Bacteriology:______________________

Other________________________________________________________________________

Figure 4. Examination chart of female wild boar reproductive organs, used in this thesis (Paper I-III).

Figure 5. No apparent blood vessels in the connection between the uterus and the mesometric ligament, indicating no previous pregnancy.

Figure 6. Apparent thickening of blood vessels in the connection between the uterus and the mesometric ligament, indicating previous pregnancy.

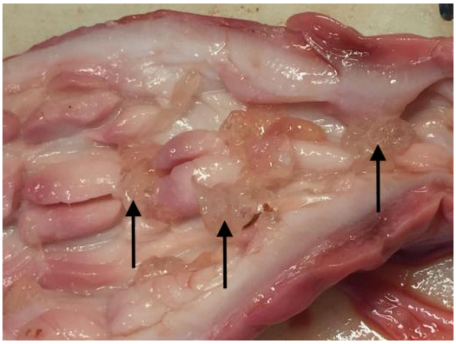

Figure 7. Black arrows pointing at remnants of ejaculate in the cervix of a wild boar.

All reproductive stages shown in Table 1 were used in Paper I. In Paper II, the animals were either recognized as pre- or post-pubertal because of the aim of that study. Animals were defined as post-pubertal if the ovaries contained one or more CL, or if the uterus showed signs of a previous pregnancy. In Paper III, animals in prooestrus, oestrus, metoestrus, and dioestrus were all classified as cyclic.

In pregnant females, from about 2.5 – 3 weeks of pregnancy (when clearly visible embryos in foetal membranes could be observed, Figure 8), the number of embryos or foetuses was counted.

Figure 8. Early pregnancy. Embryo in foetal membranes.

Litter size was defined as the total number of embryos or foetuses found in the uterus. The ratio of the total number of embryos/foetuses per total number CL, ovulation rate (OR) was calculated. The ovulation rate and litter size were used as two measurements of reproductive potential. The crown-rump length (CRL, Figure 9) of the embryos/foetuses was measured (mm). The estimated oestrous/mating month and expected month of farrowing were calculated based on the CRL of the embryos/foetuses and a gestation length of 115 days (Henry, 1968a) according to Henry (1968b).

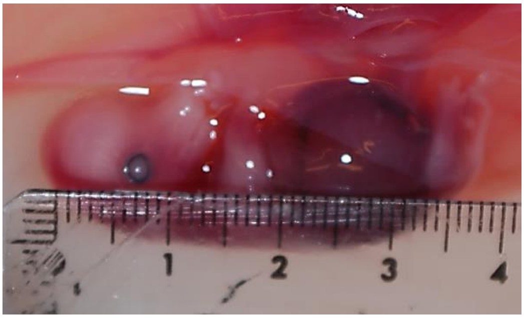


Figure 9. The crown-rump length of the embryos/foetuses was measured in order to estimate the oestrous month and the month of farrowing. The picture shows a five to six week old foetus.

Embryonic/foetal mortality, return to oestrus (appearance of both active and regressed cyclic CL), signs of disrupted oestrous cycle pattern (sizes of follicles and sizes of regressed CL not congruent), occurrence of ovarian cysts, and endometritis represent ‘abnormal findings’ (disorders). Embryonic/foetal mortality was divided into total mortality or partial mortality (entire or part of the litter dead, respectively). The term ovarian cyst (follicular or luteal cyst) was applied when cystic structures were larger than the common size of mature follicles (> 15 mm; Ebbert and Bostedt, 1993). Endometritis was recorded if signs of infection were obvious, i.e. thickening and red discolouration of the endometrium and exudate (discharge) in the uterine lumen (Figure 10).

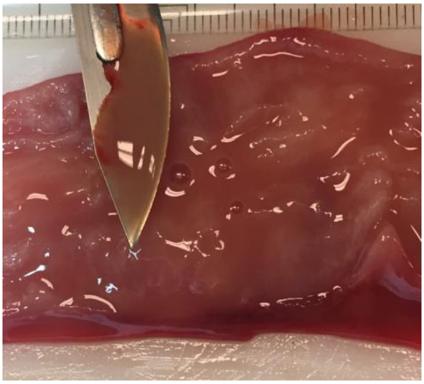

Figure 10. Endometritis; exudate in the uterus lumen.

## 3.5 Examination of stomach contents

The feed types were divided into three categories according to origin; forest material, field material and supplementary feeds. Forest material included for example acorns, leaves, needles, blueberries, and mushrooms (Figure 11a and b). Field material included for example grass, grass roots and earthworms. Supplemental feeding material constituted corn, wheat, barley, oats, beets, and silage. Supplemental feeding material was distinguished from field material according to Cellina (2008). As all samples constituted only a part of the animals` stomach contents, only the different feed types found were noted with no attempt being made to estimate the proportion in which they appeared.

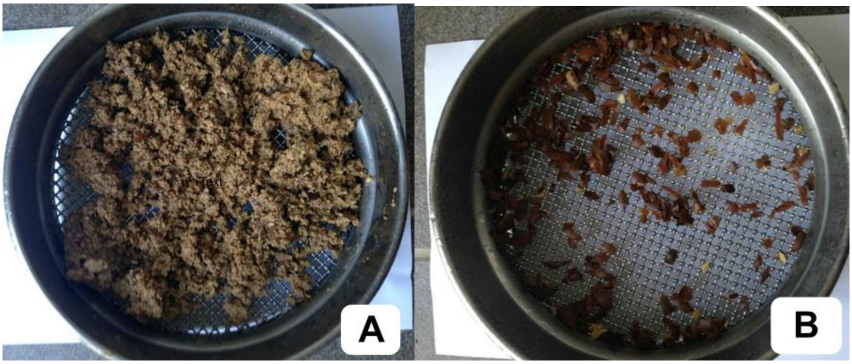


Figure 11a and b. Stomach contents (containing acorn residues) from wild boar in sieve, before (a) and after (b) being rinsed with water.

## 3.6 Reproductive pathogens and diagnostic methods (Paper IV)

There are a large number of pathogens that may affect pig reproduction, directly or indirectly. Because of time and economic limitations, a selection of pathogens was studied

First, pathogens that are known to affect pig reproduction directly or indirectly, and that are endemic in Swedish domestic pigs were selected for this study. Porcine parvovirus (PPV), Porcine circovirus type 2 (PCV-2), Swine influenza viruses (SIV), *Erysipelotrix rhusiopathiae* (*E. rhusiopathiae*), *Mycoplasma hyopneumoniae* (*M. hyo*.), and *Toxoplasma gondii* (*T. gondii*) fitted these criteria. Porcine respiratory and reproductive syndrome virus (PRRSV) *and Brucella suis* (*B. suis*) were added later because of their known effect on pig reproduction, even though they are not endemic in Sweden.

Moreover, *Mycobacterium bovis* (bTB) was included because of its effect on wild boar populations in Europe (e.g. Spain, Barasona et al., 2016). Antibodies against these pathogens have previously been detected in different wild boar populations in Europe (see Table 1, Paper IV). However, the status of most of these diseases among Swedish wild boar is unclear.

### 3.6.1 Diagnostic tests

The sera were analysed by commercial enzyme-linked immunosorbent assays (ELISA) in accordance with the manufacturers' instructions (Table 3, Paper IV). The serology was performed at Instituto de Investigación en Recursos Cinegéticos (IREC, Ciudad Real, Spain).

There are different types of ELISA, but the principle is the same for all methods, namely the use of specific antibodies against a unique antigen, thereby creating a detection signal by an enzymatic colour reaction (Figure 12).

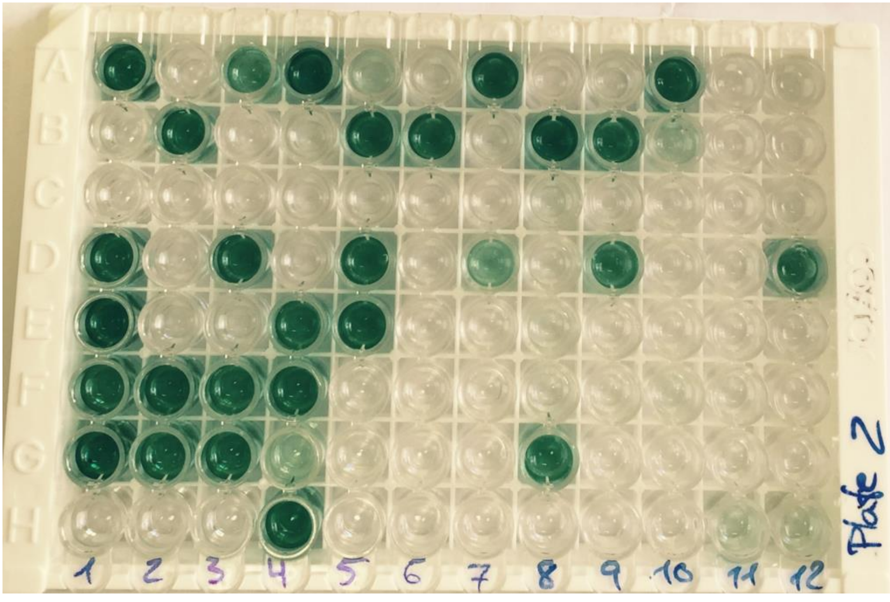


Figure 12. Microtiter plate with different coloured cells indicating positive and negative results.

## 3.7 Statistical analyses

The year was divided into four seasons; spring (March-May), summer (June-August), autumn (September-November), and winter (December-February, Paper II and III). After age determination, the animals were classified as juveniles (age < one year), yearlings (aged one to two years), and adults (age > two years). For Paper II, animals in the age-span 5 to 15 months were further sub-divided into three age groups according to the classifications of Matschke (1967); 5 to 8 months, 7.5 to 12 months, and 12 to 15 months (with an overlap between the two first age classes, 7.5 months and 8 months). However, this overlap was not considered a problem because the animals could only be grouped into one of these classes based on their tooth eruption pattern. For animals where only BW was noted, FDWs were estimated using the relationship FDW=-1.855+0.810*BW, based on the known relationship between BW and FDW of 296 animals [Lundeheim, unpublished data]. For the statistical analyses, season and age class were regarded as categorical variables, and weight as a continuous variable.

The statistical analyses were performed either in R (R Core team, 2013) or SAS (SAS Institute Inc., 2011). One-way analysis of variance (ANOVA) was used to test for differences in weight and length of the organs between and within groups formed according to their reproductive stage and sampling region (Paper I). In order to determine the effect of different explanatory variables (season, age class, weight) on the response variable, analysis of variance (general linear model or generalized linear model) were used. When the response variable was binary (such as reaching puberty, Paper II, and reproductive stage, Paper III), data were analysed using generalized linear models (see e.g. Olsson, 2002) with a binomial distribution. When the response variable was continuous (weight, OR, litter size, Paper III), a general linear model was used. Differences in proportions (such as the proportion of animals with reproductive abnormalities in various age classes and sampling regions (Paper I), and the proportion of animals that had reached puberty according to different definitions (Paper II), were analysed with Fishers' exact test. Pearsons' chi-squared test was used to analyse the sero-prevalence of pathogens in different age classes and sampling regions (Paper IV). P-values lower than 0.05 were considered as significant.

# 4 Results

## 4.1 Reproductive characteristics and abnormal findings (Paper I)

This is a summary of the key finding from the four studies. Of the 569 female wild boar included in Paper I, 129 animals were classified as in anoestrus, 103 had ovarian cyclicity (in prooestrus, oestrus, metoestrus, or dioestrus), 122 were pregnant, 204 pre-pubertal, and 11 had disturbed oestrous cycle. In 45 (34.9 %) of the anoestral animals, regressed CL of pregnancy (2 to 4 mm in diameter) were found indicating that these animals had been recently pregnant (within one-two months, Figure 13). Of these animals, only three showed signs of lactation when culled.

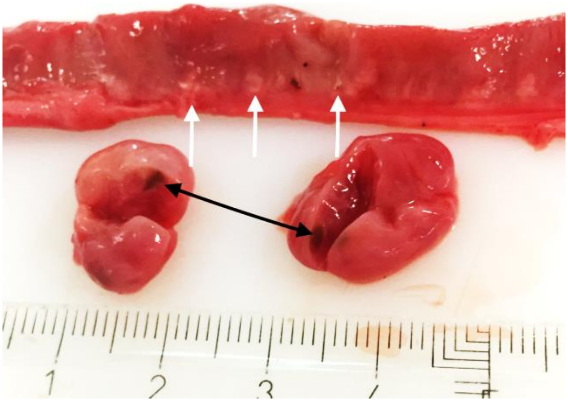


Figure 13. Ovaries and uterus from a wild boar defined as 'recently being pregnant'. Black arrows point at regressed corpora lutea of pregnancy, white arrow points at marked vessels in the connection between the uterus and the mesometric ligament.

## 4.2 Descriptive measurements and reproductive stages

The means and ranges of uterine weights and lengths in the different reproductive stages are presented in Paper 1, Table 3 (n = 436, pregnant animals and animals with disrupted ovarian cycle pattern not included). There were significant differences in the uterine weight and length among animals in anoestrus, pre-pubertal animals, and cyclic animals ($p < 0.001$). Uterine weight (within the pre-pubertal, anoestral, oestral, and dioestral animals), and length (within the pre-pubertal and anoestral animals) also varied significantly between sampling regions.

## 4.3 Abnormal findings

Reproductive abnormalities, i.e. disorders, were found in 58 (10.2 %) of all examined reproductive organs. The proportion of the abnormal findings increased with age ($p < 0.001$) and was significantly higher in the region of Uppland compared with the region of Blekinge. No other significant difference was found between regions. Embryonic/foetal mortality (Figure 4, Paper I) was found in eleven of the pregnant uteri (9.0 %, n = 122). Of these, 10 had partial and one had total embryonic mortality.

In addition, embryonic remnants were found in two other animals that were classified as having a disturbed cycle pattern. Among the cyclic and pregnant animals, (n = 225), 22 (9.8 %) had returned to oestrus (both fresh and regressed CL in the ovaries, (see Paper I, Figure 5,), and eight animals (3.6 %) had ovarian cysts, of which four (1.8 %) had a single cyst and four (1.8 %) had multiple cysts.

Endometritis was macroscopically observed in six of the examined uteri (1.6 %) of post-pubertal animals (n = 365). Parovarian cysts (remnants from embryonic development) were found in the bursa ovarica of 23 of 569 animals (4.0 %, Figure 14).

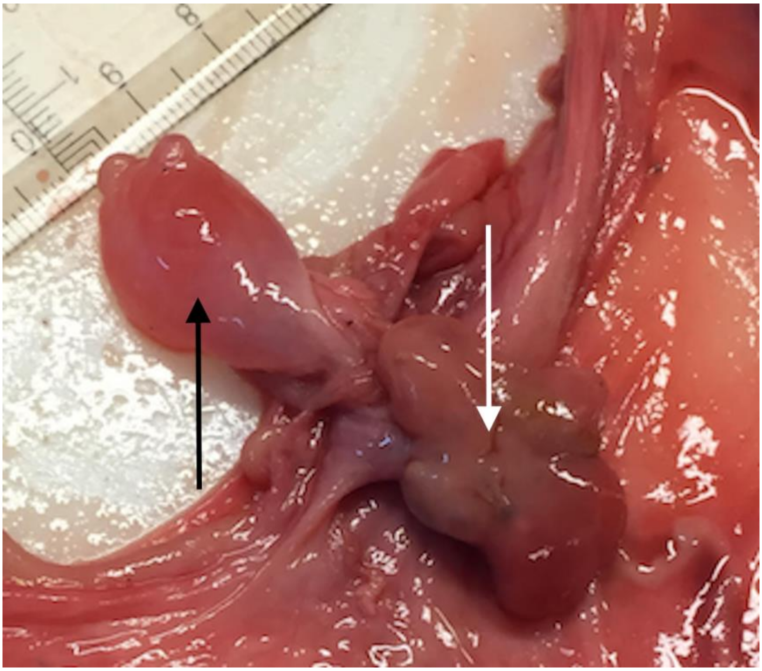

Figure 14. Parovarian cyst in a wild boar. White arrow pointing at ovary, black arrow pointing at a parovarian cyst.

No signs of other congenital anomalies were found in the organs. One animal had minor adhesions between the ovary and bursa ovarica. This animal also had two mummified foetuses (partial foetal death).

## 4.4 Puberty in female wild boar (Paper II)

In total, 29 (16.6 %) of the 175 examined animals (age between 5 and 15 months) were considered as post-pubertal. In these animals, CL were found in all except one animal, but in that one, the uterus showed signs of a previous pregnancy (marked vessels, see above). Post-pubertal females were found in all seasons apart from summer (Figure 15).

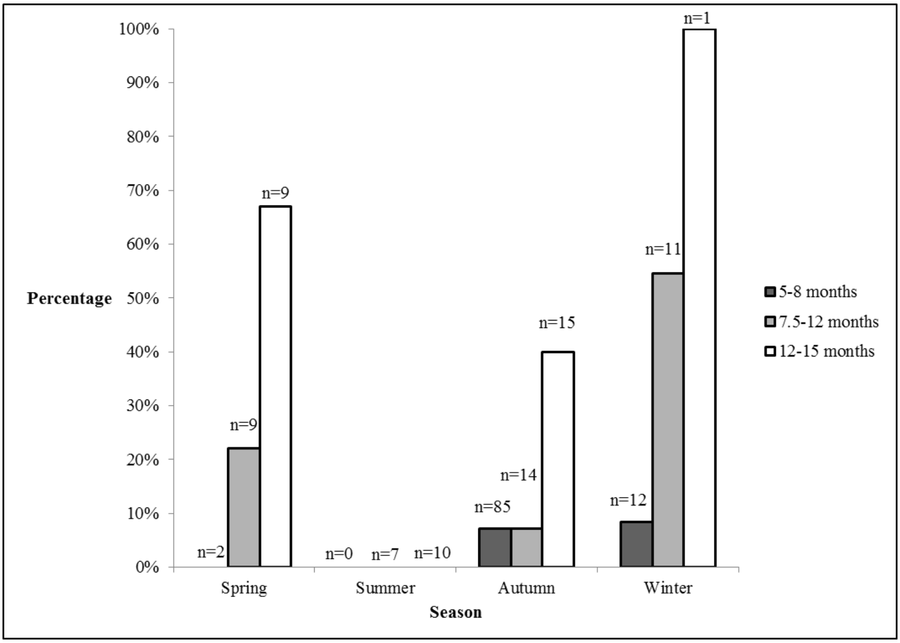


Figure 15. The proportion of post-pubertal 5-15 month old female wild boar according to age class and culling season, where 'n' shows the total number culled per age class (Paper II).

In a first step, age class, weight and season were analysed separately in relation to puberty. All of them showed significant association to puberty: (weight, p < 0.0001; season, p = 0.0079 and age class, p = 0.0006). In the joint model, season (p = 0.0395) and weight (p = 0.0002) were significantly associated with puberty, but age class (p = 0.7398) was not. There were significant differences in the proportions of post-pubertal animals and animals found with CL or follicles > 4 mm in the different age classes (p < 0.001, Table 1, Paper II).

## 4.5 Reproductive potential and seasonality (Paper III)

In total, 575 individuals met the inclusion criteria for the study in Paper III (with complete sets of reproductive organs). The number of animals in the different reproductive stages were: 207 (36.0 %) pre-pubertal, 131 (22.8 %) anoestral, 104 (18.1 %) cyclic, 123 (21.2 %) pregnant and 11 (1.9 %) had a disrupted ovarian cyclic pattern. The proportions of animals in different reproductive stages excluding pre-pubertal animals and animals with disrupted oestrous cycle, as illustrated in Figure 16, were significantly affected by season ($p < 0.0001$).

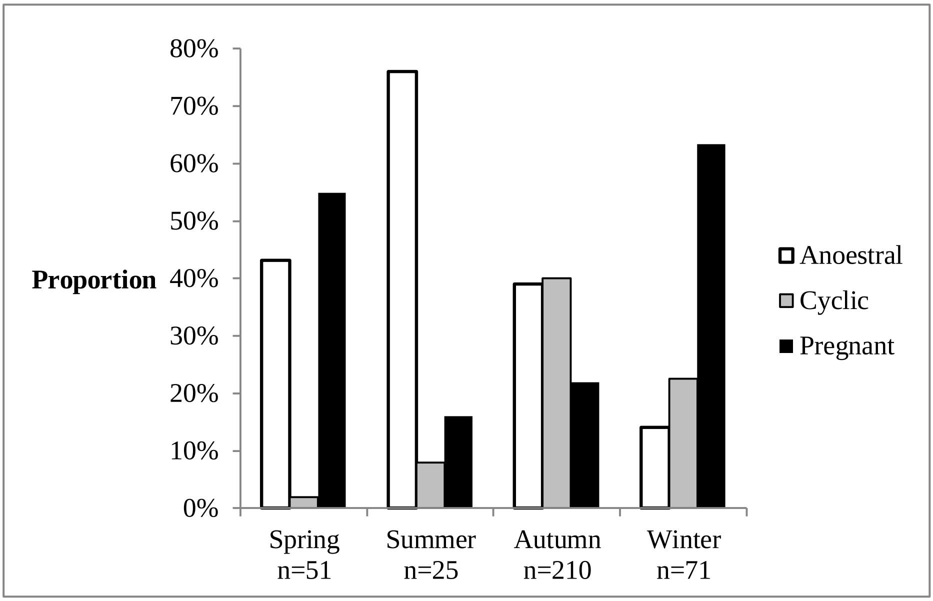


Figure 16. The proportion of female wild boar in different reproductive stages in relation to culling season (excluding pre-pubertal animals and animals with disrupted ovarian cyclic pattern, Paper III).

Pregnancy was detected in 122 animals. After excluding 11 pregnant females with embryo mortality and 10 animals in an early pregnancy state (< 2.5 weeks, Figure 17), the mean number of CL was 6.4 (range 2-11; n = 101) and the mean number of embryos/foetuses was 5.4 (range 1-9). The ratios between the number of embryos/foetuses and the number of CL for the different age classes were: 0.75 (juveniles, n = 7), 0.81 (yearlings, n = 24) and 0.85 (adult, n = 31).

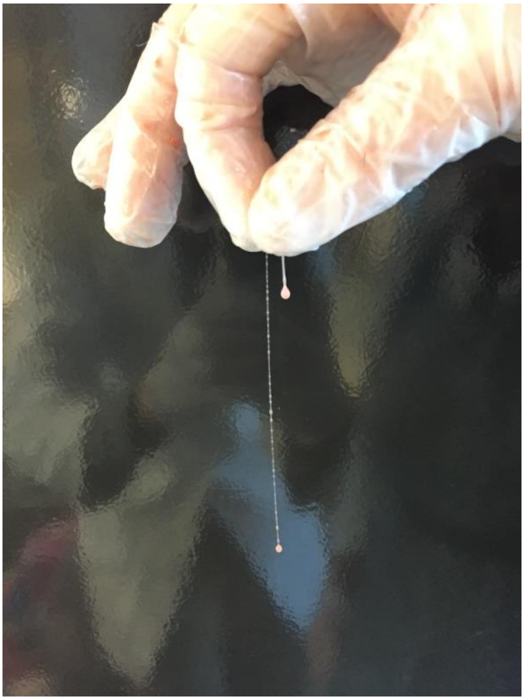

Figure 17. Foetal membrane at early pregnancy state (< 2.5 weeks).

The estimated oestrous/mating- and farrowing months based on the crown-rump length (CRL) of embryos/foetuses from 105 pregnant female wild boar sampled between November 2012 and December 2014, are illustrated in Figure 18.

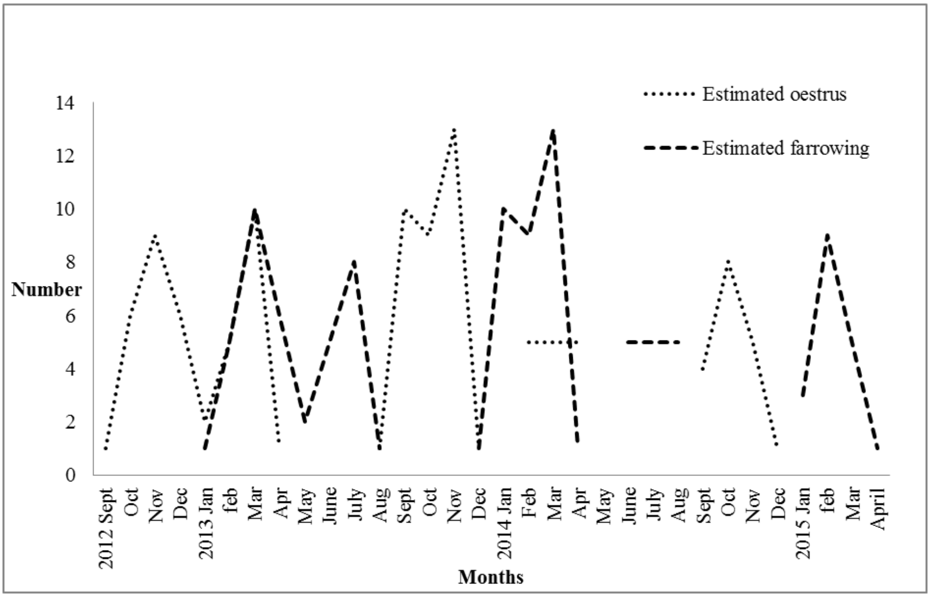


Figure 18. The estimated oestrous/mating- and farrowing months based on the Crown-Rump-Length (CRL) of embryos/foetuses from 105 pregnant wild boar (Paper III).

The reproductive potential (here defined as the number of CL or intra uterine litter size) as well as pregnancy rate among the sampled wild boar females (n = 442, Table 3, Paper III) increased significantly with age class ($p < 0.001$).

## 4.6 Stomach contents

Examination of the stomach contents was performed in 222 of the sampled animals. Of these wild boar, 57 were sampled in spring, 41 in summer, 86 in autumn, and 38 during winter. Supplemental feeding material was found in a high proportion of the wild boar in all seasons. The proportion of feed type per culling season is presented in Figure 19.

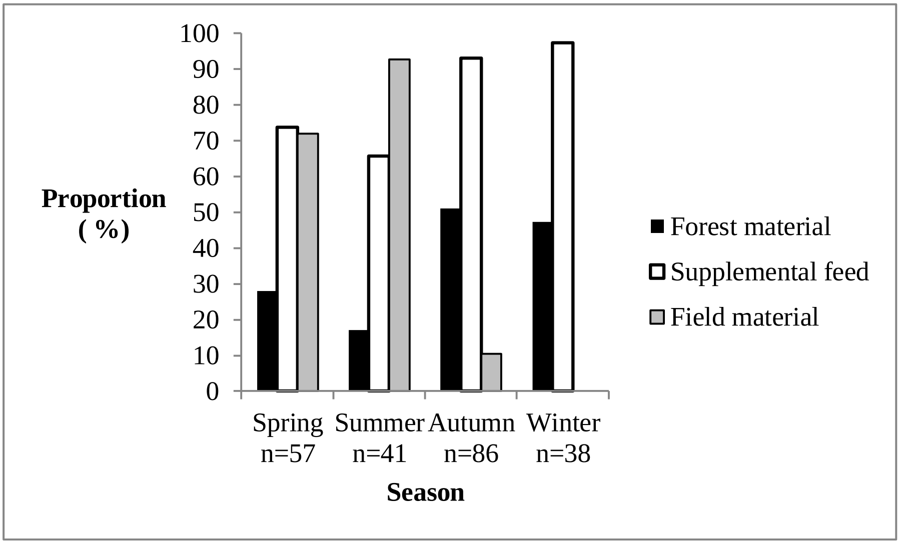


Figure 19. The proportion of wild boar (n = 222) with the respective feed type (forest material, supplementary feed, field material) found in their stomach contents and the distribution according to culling season. The n-values show the number of animals examined per season.

## 4.7 Sero-prevalence (Paper IV)

The distribution of age class, sampling region, number of serologically tested animals and sero-prevalence are presented in Paper IV, Table 4. Seventy-eight percent of the samples were positive for PPV, 99 % for PCV-2, 3.8 % for SIV, 17.5 % for *E. rhusiopathiae*, 24.8 % for *M. hyo*., and 28.6 % for *T. gondii*. No antibodies were detected against PRRSV, *B. suis*, or bTB in the sera.

Age class was positively correlated to the prevalence of antibodies against PPV ($p < 0.05$), SIV ($p < 0.05$), *M. hyo*. ($p < 0.05$), *E. rhusiopathiae* ($p < 0.05$) and *T. gondii* ($p < 0.05$). The antibody prevalence increased with the individual's age for PPV and *T. gondii* but for *E. rhusiopathiae* it was only higher in adults when compared with juveniles. For SIV the highest antibody prevalence was observed in juveniles, whereas for *M. hyo*. yearlings displayed the highest exposure. In contrast, animal age class was not associated with PCV-2 antibody prevalence.

Sampling region was only significantly associated with the prevalence of antibodies against PPV and *M. hyo*. The antibody prevalence of PPV was higher in Södermanland (Figure 3) whereas the prevalence of antibodies against *M. hyo*. was higher in Blekinge compared to the other regions.

# 5 General discussion

This thesis describes some aspects on the reproduction of female wild boar in four Swedish regions. The aim of this work was to provide new knowledge about wild boar reproduction under Swedish conditions, such as the characteristics of reproductive organs, occurrence of abnormal findings, puberty in wild boar gilts, reproductive seasonality and potential, as well as the sero-prevalence of reproductive pathogens. The hope is that the information given in this thesis will help authorities, land owners and wildlife managers in their decision-making when managing the species.

## 5.1 Characteristics of reproductive organs

Knowledge about the normal characteristics of the reproductive organs is necessary when making comparisons between populations of game species but also, as in this case, when comparing a wildlife species with a related, domesticated species. In addition, one needs to know what is normal before abnormalities can be recognized. In Paper I, the reproductive organs of Swedish female wild boar were examined post-mortem, and the reproductive stage determined. The relation between both weight and length of the organs and reproductive stage were in accordance with studies of domestic pigs by Heinonen et al. (1998) and Tummaruk and Kesdangsakonwut (2014). One explanation for this observation is that the uterus, i.e. the endometrium, mirrors the production of steroid hormones in the ovaries. Within the different reproductive stages, a substantial variation in the ranges of uterine weight and length was found. In addition, the mean uterine weight and length differed significantly between sampling regions. The cause of these differences remains unknown but the uteri from domestic sows and gilts are generally heavier (Dalin et al. 1997) and longer compared to those of wild boar. Preliminary data (Malmsten et al., unpublished) suggest that there are genetic differences between the studied wild boar populations (Figure 20) as well as signs of introgression of domestic pig (Figure 21). Thus, these genetic variations might explain some of the phenotypic variations. Further studies to elucidate these relationships will be performed. In domestic pigs, the length of the uterus may influence reproductive output, i.e. a longer uterus has the capacity to accommodate larger litters (Wu et al. 1987).

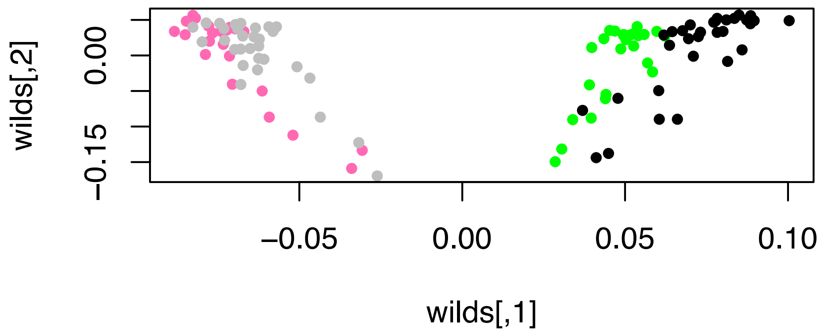


Figure 20. Principal component analysis (PCA) for the 80 K SNPs data set for 108 wild boar, from four Swedish regions, delineates the animals into two main genomic clusters in accordance to their geographical origin; geographically closer populations are clustered in the PCA plot. (Grey = Skåne, Pink = Blekinge, Green = Södermanland, Black = Uppland).

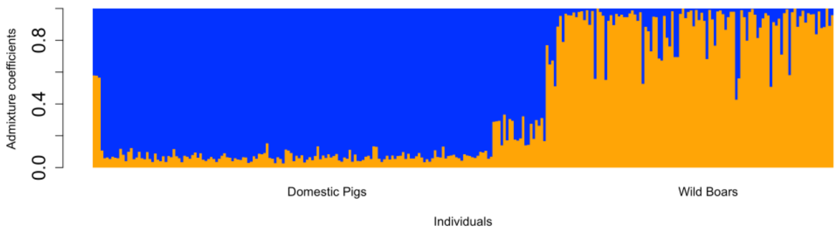


Figure 21. Bayesian ancestry inference using the R package LEA (K=2, Frichot and François, 2015) showing the differentiation of the wild boar (n = 108) from domestic pigs (n = 170) with signs of introgression in a few of the individuals.

## 5.2 Abnormalities of the female reproductive organs

Most of the abnormal findings detected (Paper I) were signs of reproductive disturbances (return to oestrus, disturbed oestrous cycle, endometritis and embryo/foetal mortality) which would have affected its reproductive performance if the animal were still alive. The prevalence of abnormalities increased significantly with age, which was as expected. Older females have usually experienced reproductive activity such as pregnancies and farrowing and may thus have been subjected to some kind of disturbance. The regional variation found in this study may result from a varying occurrence of infectious disease, as well as differences in population density, and/or skewed sex ratios.

Due to lack of similar studies on wild boar reproduction, the present findings are mainly discussed in relation to post-mortem studies of reproductive organs from domestic pigs. However, in most studies on domestic pigs, the organs were obtained from gilts and sows culled due to poor reproductive performance, which likely increases the reported frequency of reproductive disorders.

The ratio between the number of embryos/foetuses and the number of CL in this study varied from 0.75 in juveniles up to 0.85 in adults. Comparable numbers from a Spanish study are 0.85 and 0.95. In the same study (Ruiz-Fons et al., 2006), the ovulation rate and litter size were smaller compared to the present results. It is possible that the ratio between CL and number of embryos/foetuses decreases with ovulation rate. In domestic pigs, the ovulation rate is higher than in wild boar. The ovulation rate in commercial domestic sows is nowadays normally 20 to 30, with an embryonic survival of about 60-70 % at 30 days of pregnancy, decreasing to approximately 40-60 % at 50 days (the lower figure being for the higher parity number, see Foxcroft et al., 2009).

Only macroscopically evident embryonic/foetal mortality was considered in this study. It is not possible to observe embryonic mortality macroscopically earlier than two weeks of pregnancy. However, return to oestrus or a disrupted ovarian cycle pattern may be signs of previous embryonic mortality. Embryonic survival in domestic pigs is affected by nutrition (Almeida et al. 2000; Condous et al. 2014; Langendijk and Peltoniemi, 2013; Zak et al. 1997), epigenetic variance (Vinsky et al. 2007), pathogens such as porcine parvovirus (PPV, Mengeling et al. 2000), and porcine circovirus type 2 (PCV-2, Mateusen et al. 2007), maternal stress (Brandt et al. 2007), and embryo-maternal communication (see Roberts et al. 1993; Østrup et al. 2011). The wild boar included in the present study had access to supplementary feed; therefore shortage of feed or malnutrition are unlikely to be explanations for the observed embryonic mortality. In one of the pregnant females, mummified foetuses were found. Unfortunately, serum from this animal was not included in the serological study, but the presence of mummified foetuses indicates infectious disease as a plausible cause. The high prevalence of some pathogens, such as PPV and PCV-2, found in Paper IV, strengthens this hypothesis.

The main hunting period of wild boar in Sweden occurs during autumn and winter. During autumn (Oct–Nov) most of the post-pubertal female wild boar will show oestrus, be mated, and become pregnant as presented here. It cannot be excluded that frequent driven hunts with dogs and beaters during this period would stress pregnant females. Stress may affect the early reproductive process and lead to failure in fertilization and embryonic development, as shown in domestic pigs (see review by Einarsson et al. 2008).

In the wild boar studied, 9.8 % of 225 animals (excluding pre-pubertal and anoestral animals) had returned to oestrus. In domestic pigs in Sweden, return to oestrus/not pregnant is the most common reason for culling gilts or sows (Dalin et al. 1997; Engblom et al. 1997). Return to oestrus can, as mentioned above, result from an embryonic death at an early stage. However, in wild boar ”not mated” may also be a reason. In Sweden, mature male wild boar are hunted for trophies, whereas there is low hunting pressure on adult females/sows, which may lead to skewed sex-ratios. There are no data on population compositions for wild boar in Sweden. A strongly skewed sex ratio may, however, result in a shortage of males with females in oestrus not being mated, and consequently returning to oestrus.

Porcine ovarian cysts may be classified as single or multiple cysts (Miller, 1984; Wrathall, 1980). In domestic pigs, multiple follicular cysts cause infertility (Dalin et al. 1997), whereas a single ovarian cyst will not negatively influence reproduction. The prevalence of cystic ovaries found in this study (1.8 % for both single and multiple cysts, respectively) is lower compared with domestic pigs (16 % cystic ovaries, Ebbert and Bostedt, 1993; 14 % multiple cysts, Dalin et al. 1997; 3.1 % single and 3.1 % multiple cysts, Heinonen et al. 1998; 5.0 % single and 5.5 % multiple cysts, Tummaruk et al. 2009). As mentioned before, the higher prevalence found in domestic pigs may, however, result from the availability of examined material since domestic gilts and sows were likely culled due to poor reproductive performance.

Parovarian cysts are, in most cases, not considered to impair fertility (Heinonen et al. 1998). Einarsson and Gustafsson (1970), on the other hand, claimed that parovarian cysts may cause reduced fertility due to impaired function of the infundibulum during ovulation.

No other congenital abnormalities were observed in the examined animals. This is interesting because in domestic pigs, findings of congenital abnormalities such as hermaphroditism, *uterus unicornis,* segmental aplasia and double cervix, are frequently described (Einarsson and Gustafsson, 1970;

Dalin et al. 1997; Tummaruk et al. 2009; Tummaruk and Kesdangsakonwut, 2014).

## 5.3 Puberty in wild boar gilts

As expected, this study (Paper II) shows that the weight of the female wild boar, as well as the season, influenced the proportion of post-pubertal females. Body weight has been shown to affect age at puberty in various species (Love et al., 1993; Malmsten et al., 2014), including wild boar (Gaillard and Jullien, 1993). Females of most mammalian species need to pass a certain threshold for body mass to be able to reproduce. Female pigs that pass this threshold early, due to favourable conditions such as high feed availability leading to fat deposition (Barb et al., 2005), will start reproducing at a young age. A minimum amount of body fat seems to be needed for the onset of puberty. Furthermore, adipose tissue and its production of leptin are also important for pubertal development in pigs (Hausman et al., 2012).

In wild boar, as in many wildlife species, population growth is dependent on the proportion of females that are able to reproduce. In a species where reproductive capability is reached at young age, the population growth may be fast, especially in pluriparous species. Only 11.4 % of the animals aged < 12 months were classified as post-pubertal in the present study. In the southern and middle part of Sweden, feed availability (regardless of supplementary feeding) is probably high for wild boar, in the form of crops and natural feed. During periods of natural feed shortage, such as in winter, supplementary feeding is extensive. Considering food availability, one would expect that the proportion of post-pubertal animals would be higher in the present study and more in accordance to other studies (Ahmad et al., 1995; Cellina, 2008) using the same definition of puberty i.e. the occurrence of ovarian CL. Extensive supplementary feeding has been suggested to be one of the main causes of reaching puberty at a young age (Cellina, 2008). Cellina (2008) also reported that 24 % of supplementary-fed wild boar females aged less than 12 months were post-pubertal in Luxemburg, and the youngest was four months old. No females younger than five months of age were investigated in the present study. Based on the low proportion (7.1 %) of post-pubertal gilts aged 5-8 months, the proportion of even younger females that had reached puberty is probably negligible. However, due to the wide age category range (months), it is not possible to confirm whether pregnant animals in this study had reached puberty within the noted age category, or at a younger age. Domestic crossbred pigs normally reach puberty at 6 to 7 months of age (Hughes, 1982; Dalin and

Elisasson, 1987), and gilts that not reached puberty at the age of eight months are considered to have delayed puberty (Dalin and Eliasson, 1987).

A study in Pakistan (Ahmad et al. 1995) showed a high proportion (25/32, 78 %) of wild boar gilts aged 4 to 7 months with CL, which differs from the animals in this study as well as from a study in Luxemburg (Cellina, 2008). This indicates that the genetic background of wild boar may also be of importance for the pubertal age as seen in the domestic pigs (Hutchens et al., 1982). The topic requires investigation in regional/international studies.

Different definitions of puberty will affect the proportion of female wild boar gilts considered to have attained puberty in a population as well as in different age classes. In the present study, 79.8 % of the wild boar aged 5 to 8 months were found with follicles ≥ 4 mm or with CL, and would have reached puberty according to the definition used by Gethöffer et al. (2007). The latter authors described the probability of juveniles having attained sexual maturity at an age of eight months to be 80 %. However, large numbers of follicles will become atretic (Dalin, 1987), even before puberty. Thus, using follicles of a specific size to define puberty may result in an overestimation of the proportion of wild boar females considered being capable of reproducing. Using the definition of puberty as ‘presence of ovarian CL’, as in this study, means that ovulation has indeed occurred, which is a prerequisite for a fertile female. This is the definition of puberty used for domestic pigs (Dalin and Eliasson, 1987; Eliasson et al., 1991) and other domestic species when examining reproductive organs.

## 5.4 Reproductive seasonality

Although the majority of the female wild boar showed reproductive seasonality (Paper II-III) in accordance to previous studies (Mauget, 1982; Ježek et al., 2011), cyclic and pregnant females were found in all seasons. This pattern was confirmed by the estimated oestrous/mating and farrowing months based on the CRL of embryos/foetuses. These observations show that farrowing may occur ‘off-season’ in Sweden, as also observed in other studies when feed is accessible throughout the year (Fonseca et al., 2004; Bieber and Ruf, 2005). Despite the lack of samples from some months, two distinct peak periods for oestrus/mating (one in November and one in March) were seen in the material from 2013, and were also evident, but not as pronounced, in 2014 (Figure 18). This would have resulted in peaks of farrowing in March and July, if the animals had still been alive. Mating in March is not considered normal for short-day breeders, such as wild boar. However, this bimodal distribution of birth has been described previously in wild boar (Mauget, 1972; Mauget, 1982; Ježek et al., 2011). The second peak may be explained to result from one ore more of the following: 1) some factor(s) that make an early spring litter die, with the result that the sow goes into oestrus again and becomes pregnant; 2) young females taking part in/entering reproduction during spring, i.e. juveniles that had not reached the necessary weight in the autumn mating season; 3) genetic influences of domestic pigs in the wild boar population that affect the seasonality of reproduction (Mauget, 1972). Factors behind the first explanation may be harsh weather conditions, scarcity of feed, predation or diseases that affect litter survival. In the macroscopic examination of the reproductive organs a large proportion of animals in anoestrus with regressed CL of pregnancy were observed (Paper I and III). This suggests that these animals had recently been pregnant. In practice, this means that they had either a) lost their piglets shortly after birth due to piglet death (because of the factors mentioned above), or had deserted their piglets because of an unknown cause; b) had aborted at a late stage, i.e. lost the litter before birth due to poor body condition or disease; or c) were shot despite having a litter (although this eventuality is unlikely since hunters had neither seen piglets nor reported sows with drawn teats). The studied populations had access to supplementary feed all year round, with the result that scarcity of feed and subsequent starvation as explanations to the lost litters and second birth peak are less likely. The observed occurrence of infectious pathogens may have contributed to lost litters and the second birth peak. Another potential cause to the second birth peak is the second explanation given above, i.e. young females taking part in reproduction during spring. In the present study, the female wild boar involved in the second birth peak (June-July) had a mean live weight of 54.3 kg (n = 13, range 34-70.6 kg), However, these animals were not age-determined which would have been needed in order to draw such a conclusion. The third explanation, genetic influence of domestic pigs (considered as being poly-oestral throughout the year if not pregnant), should not be excluded as we have found signs of introgression of domestic pig in the studied wild boar populations (Malmsten et al., unpublished; Figure 21).

## 5.5 Reproductive potential

The reproductive potential indicated by ovulation rates (CL) and litter size of the wild boar studied was high (6.4 and 5.4, respectively, (Paper III) compared to litter sizes reported from other countries, e.g. Italy 5.0 (Boitani et al., 1995), Switzerland 4.8 (Moretti, 1995), Iberian Peninsula 3.6 (Fernández-Llario and

Mateos-Quesada, 1998), and Portugal 4.2 (Fonseca et al., 2004), but not as high as in Germany; 6.6 (Frauendorf et al., 2016). The reproductive potential of wild boar can be affected by many factors, such as feed availability, climate (Moretti, 1995; Fernández-Llario and Carranza, 2000; Geisser and Reyer 2005; Servanty et al., 2007), the genetic background including influence of domestic pigs (Mauget, 1972; Booth, 1995; Gongora et al., 2003), and the weight and age of gilts and sows (Mauget, 1982; Fernández-Llario and Carranza, 2000). High natural feed availability (Massei et al., 1996), as well as the availability of supplementary feed (Fernández-Llario and Mateos-Quesada, 1998; Fernández-Llario and Carranza, 2000), may increase litter sizes. In Sweden, although controversial, supplementary feeding is applied to a varying extent throughout the wild boar range. At the same time, the availability of natural feed in Sweden is probably high for wild boar, both in forested and agricultural habitats, especially in the southern and central parts of Sweden. During the winter periods when natural feed is less abundant (e.g. no crops *et cetera* available), supplementary feeding is important for wild boar feed consumption, as shown in the present result.

In contrast to southern and central Europe (Mauget, 1978), Swedish summers are not likely to constitute a regulating season for the wild boar. Instead, the summer conditions in southern and central Sweden normally result in a long vegetation period without high ambient temperatures or drought (SMHI.se, 2016). Such conditions are favourable for wild boar (Fernández-Llario and Carranza, 2000; Frauendorf et al., 2016) and possibly contribute to the high reproductive potential, found in Paper III.

The genetic introgression of domestic pigs (Figure 21-22) in the wild boar populations may also have influenced the reproductive potential, as previously described (Booth, 1995; Gongora et al., 2003).

Figure 22. Different phenotypes (coat colour and pattern) of wild boar foetuses, from the same litter, may be signs of genetic introgression of domestic pigs.

In agreement with previous studies (Boitani et al., 1995; Fernández-Llario and Mateos-Quesada, 1998; Fonseca et al., 2004; Santos et al., 2006; Frauendorf et al., 2016), pregnancy rate and reproductive potential increased with age and weight. The Swedish hunting regulations (Jaktförordning, 1987:905) prohibit culling of females with piglets, and large females in general (even those without piglets) are often voluntarily banned from culling by hunters. Animals included in this study, therefore, cannot be considered as a random sample of the whole population. Instead, the true proportion of large, adult females may have been higher in the population than in the studied material, and the average reproductive potential presented here may thus be underestimated.

## 5.6 Sero-prevalence of selected reproductive pathogens

It has been shown that promoting high wild boar densities, mainly for hunting purposes, also increases prevalence and transmission of pathogens (Vicente et al., 2004). However, the extent to which high wild boar densities are counterproductive because of the indirect effects caused by pathogens in a population are unknown and may vary with the type of pathogen. Knowledge about pathogens circulating among wildlife, such as the wild boar, is of major importance not only for management of the species themselves, but also for domestic animals. For example, to prevent transmission of diseases from wildlife to domestic animals, it is necessary to know the reservoir species of the pathogens. In addition, some pathogens also have zoonotic potential, and therefore the risk of transmission to humans should not be neglected (see Ruiz-Fons 2015). In this thesis (Paper IV), no antibodies against PRRSV, *B. suis*, or bTB were detected in the sera of wild boar. This was expected because Sweden is reported to be free from these pathogens in domestic animals (SVA, 2015). On the other hand, this study presents high prevalence of antibodies against PPV, PCV-2, *E. rhusiopathiae*, and *M. hyo*. in the wild boar populations. This highlights the potential of the wild boar as a reservoir of different pathogens that can affect domestic pigs (or vice versa), and in some cases such as *E. rhusiopathiae* and *T. gondii,* be transmitted to humans.

The sero-prevalence of pathogens detected are within the same range (SIV, *E. rhusiopathiae*, and *M. hyo*.) or among the highest ever reported (PPV and PCV-2,) in wild boar populations in Europe (Paper IV, Table 1). Previous studies have shown that intensive management of wild boar populations, such as fencing and feeding, can have a profound effect on the prevalence of pathogens (Vicente et al 2004; Ruiz-Fons et al. 2006; Hälli et al., 2012). Even though the wild boar in Sweden is free-ranging (i.e. non-fenced), population densities are high in some areas, and comparable to those seen in fenced hunting areas in Spain (Acevedo et al., 2007). Supplementary feeding likely contributes to these high densities. The common feeding system also results in aggregations of animals at the feeding sites, in turn leading to increased contact between individuals and groups and thus increasing the potential for pathogen transmission. High wild boar densities also enhance the risk of spreading other severe diseases, such as African swine fever virus (ASFV), if it enters Sweden from neighbouring countries (Estonia, Latvia, Lithuania and Poland) where it is present (European Commission, 2017).

Currently, there is a gap of knowledge concerning the direct effect of the pathogens detected in this study on wild boar, both at individual- and population levels. Studies of clinical manifestation of diseases in free ranging wild boar are few. However, clinical cases of PPV (Zhang et al., 2010), PCV-2, *T. gondii*, and acute septicemic erysipelas in farmed wild boar have been reported (Yamamoto et al., 1999; Risco et al., 2011). In Sweden, sporadic cases of PCV-2 and PPV have been diagnosed by the national wildlife disease surveillance program at the National Veterinary Institute (SVA) [personal communication C. Bröjer]. This shows that wild boar are susceptible to these pathogens and can develop severe disease which, if widespread in a population, may affect population dynamics and health. More studies on the direct effect of these pathogens on wild boar are needed to confirm this scenario.

## 5.7 Management implications

Wild boar management is complex and time consuming, and should ideally be adapted to annual variations in the environment, as well as regional preconditions. This requires repeated data collection and improved knowledge of wild boar population biology, to which this thesis contributes. When it comes to management of a game species, a proper management plan, including formulation of methods for how to decrease the population, keep it stable, or increase the number of animals, is needed. These different goals may vary between years. High densities of wild boar may be desirable to many hunters for several reasons, e.g. economic (hunting tourism), recreational, and also availability of game meat. On the other hand, high densities increase the risk for, and extent of, agricultural damage and may also contribute to the spread and/or maintained circulation of certain pathogens. The results of this thesis suggest that both the reproductive potential and the pregnancy rate of the sampled populations are high compared to other European wild boar populations. These levels probably result from the combined effects a generally favourable climate, available feed resources and possibly also the genetic background of the populations. Both the reproductive potential and pregnancy rate increase with the age of the female. A management plan designed for areas where the intention is to decrease the wild boar population, should thus allow culling of old/heavy female wild boar and, preferably, limiting feed resources. However, culling large females is controversial in Sweden. Even though permitted by current hunting legislations, wildlife managers and hunting teams often prohibit shooting of all large females, even those without dependent piglets. Sometimes shooting a large female result in the imposition of a fee to be paid to the hunting estate/team. The argument for this is that it would minimize the risk of shooting females with dependent piglet who are protected by Swedish hunting legislation. Even though large females were protected from culling in the study areas, almost 20 % of the weight-determined animals (n = 578) in this material had a body weight exceeding 80 kg. Of these, 90 % were shot during driven hunts in the main hunting season. The reason for these 'mistakes' could either be that the hunters thought that the females were males or that they seemed to be smaller than they actually were. This indicates the difficulties in distinguishing males and females, and to estimate the body weight in a hunting situation. However, when examining these 'mistakes' (animals with a BW ≥ 80 kg) in detail, only two animals were lactating when culled and thus can be considered as being wrongfully shot according to current legislation. The low frequency of wrongfully shot animals suggests that most of them actually were without dependent piglets, indicating a seasonal reproductive pattern. If all of them had been perfectly seasonal, the ethical issue described above would not have been a problem. However, because of females farrowing ‘off-season’, there is always a risk that a female has dependent piglets. According to the present results, if adult females are to be harvested, the farrowing pattern (Figure 18) shows that the risk of them having nursing piglets is low in October-November, although this could vary between years (Fonseca et al., 2004, Bieber and Ruf, 2005). The low frequency of wrongfully shot animals may also suggest that although body weight and sex determination is difficult, Swedish hunters are skilled in determining whether a female wild boar is likely to have piglets or not, which is also assumed by both legislation and hunting ethics.

# 6 Conclusions

This thesis provides new information on the basic reproductive parameters of female wild boar in Sweden. The specific conclusions drawn from this thesis are as follows:

- The weight and length of the uterus mirrors the present reproductive stage
- In the studied wild boar populations, approximately 10 % had some sort of reproductive abnormalities, which could affect the reproductive outcome/performance.
- A low proportion (compared to previous European studies) of the examined young animals, aged between five and 15 months, had attained puberty even though the food availability, including supplementary feeding, was high.
- Different definitions of puberty will result in various outcomes in the proportion of animals considered to be post-pubertal. This highlights the importance of using an adequate definition of puberty.
- The majority of the female wild boar showed reproductive seasonality. However, farrowing may occur 'off-season' which complicates wild boar hunting and may thus confer difficulties in meeting management goals.
- The reproductive potential in the studied wild boar populations was high compared to other countries. These levels probably result from the combined effects a generally favourable climate and available feed resources. The possible effect of introgression of domestic pigs should not be neglected as a contributing factor to the high reproductive potential and altered reproductive seasonality.
- The reproductive potential as well as the pregnancy rate increased with female age and weight. Therefore, if the intention is to effectively decrease the wild boar population, adult sows should be targeted for hunting, which however raises a number of ethical questions.
- The high sero-prevalence of PPV, PCV-2, *E. rhusiopathiae*, and *M. hyo*. found could be an effect of high population density and aggregation of

animals at and around feeding sites. The result demonstrates that the wild boar could serve as reservoirs of pathogens that can affect domestic pigs in Sweden.

# 7 Future considerations

The knowledge gained from this thesis raises a number of questions that should be considered in the future. First, in this thesis different reproductive failures have been described, but their causes and effects remain to be investigated. In addition, this thesis presents high prevalence of pathogens in the wild boar populations. The association between reproductive abnormalities (and other health effects) and the sero-prevalence presented will be investigated. Moreover, the role of feeding sites for the spread of infections may also be of importance and warrants further study. The genetic analyses that have been made within this project (but outside this thesis) have mainly investigated the genetic relationship between geographically different wild boar populations and the occurrence of introgression of domestic pigs. To be able to make better conclusions and predictions regarding the genetic influences on wild boar reproduction, further analyses will include associations between level of introgression and reproductive potential, ovarian cyclic pattern, and body weight. Although the reproductive potential may be comparatively high in the studied wild boar, litter survival rates, which are as crucial (or even more so), for population development are still not well described or understood. Based on previous studies, such data are not easily obtained because previously used field methods have induced negative effects on litter survival (sows leaving the litter because of disturbance *et cetera*). New methods should be developed and tested, for example using camera traps. The social role of the dominant sow (in a group) is frequently debated in Sweden, but seems to be of lesser concern in other countries. It would be valuable, e.g. for managers, to investigate group behaviour after removal of a dominant sow. Would it make younger animals more prone to feed in agricultural land, and produce more litters 'off season' if the dominant sow is removed? – These are examples of effects that have been proposed, however still only assumptions with no scientific confirmation.

# 8 References


Acevedo, P., Vicente, J., Höfle, U., Cassinello, J., Ruiz-Fons, F. & Gortázar, C. (2007). Estimation of European wild boar relative abundance and aggregation: a novel method in epidemiological risk assessment. *Epidemiology & Infection*, 135, pp 519-527.

Ahmad, E., Brooks, J.E., Hussain, I. & Khan, M.H. (1995). Reproduction in Eurasian wild boar in central Punjab Pakistan. *Acta Theriologica*, 40, pp 163-173.

Almeida, F.R., Kirkwood, R.N., Aherne, F.X. & Foxcroft, G.R. (2000). Consequences of different patterns of feed intake during the oestrous cycle in gilts on subsequent fertility. *Journal of Animal Science,* 78, pp 1556-63.

Andrzejewski, R. & Jezierski, W. (1978). Management of the wild boar population and its effect on commercial land. *Acta Theriologica*, 23, pp 309-333.

Barasona, J.A., Acevedo, P., Diez-Delgado, I., Queiros, J., Carrasco-García, R., Gortázar, C. & Vicente, J. (2016). Tuberculosis-associated death among adult wild boar, Spain, 2009–2014. *Emerging Infectious Diseases*, 22. Doi:10.3201/eid2212.160677.

Barb, C.R., Hausman, G.J. & Czaja, K. (2005). Leptin: a metabolic signal affecting central regulation of reproduction in the pig. *Domestic Animal Endocrinology*, 29, pp 186-192. Doi:10.1016/j.domaniend.2005.02.024

Bieber, C., & Ruf, T. (2005). Population dynamics in wild boar *Sus scrofa*: Ecology, elasticity of growth rate and implications for management of pulsed resource consumers. *Journal of Applied Ecology*, 42, pp 1203 – 1213.

Boitani, L., Trapanese, P. & Mattei, L. (1995). Demographic patterns of a wild boar (*Sus scrofa* L.) population in Tuscany, Italy. *IBEX Journal of Mountain Ecology,* 3, pp 197-201.

Booth, WD. (1995). Wild boar farming in the United Kingdom. *IBEX Journal of Mountain Ecology*, 3, pp 245–8.

Brandt, Y., Madej, A., Rodriguez-Martinez, H. & Einarsson, S. (2007). Effects of exogenous ACTH during oestrus on early embryo development and oviductal transport. *Reproduction in Domestic Animals*, 42, pp 118-125.

Castagna, C.D., Peixoto, C.H., Bortolozzo, F.P., Wentz, I., Neto, G.B. & Ruschel, F. (2004). Ovarian cysts and their consequences on the reproductive performance of swine herds. *Animal Reproduction Science*, 81, pp 115-123.

Cellina, S. (2008). *Effects of supplemental feeding on the body condition and reproductive state of wild boar Sus scrofa in Luxembourg*. Dissertation, University of Sussex. Doi:10.13140/2.1.5077.6642.

Condous, P.C., Kirkwood, R.N. & van Wettere, W. (2014). The effect of pre- and post-mating dietary restriction on embryonic survival in gilts. *Animal Reproduction Science*, 148, pp 130-136.

Dalin, A.-M. & Eliasson, L. (1987). Clinical, morphological and endocrinological studies in gilts with delayed puberty. *Acta Veterinaria Scandinavica*, 28, pp 263–69.

Dalin, A.-M. (1987). Ovarian follicular activitiy during the luteal phase in gilts. *Zentralblatt für Veterinärmedizin. Reihe A*. 34, pp 592-601. Doi:10.1111/j.1439-0442.1987.tb00320.x

Dalin, A.-M., Gidlund, K. & Eliasson-Selling, L. (1997). Post-mortem examination of genital organs from sows with reproductive disturbances in a sow-pool. *Acta Veterinaria Scandinavica*, 38, pp 253- 262.

Dardaillon, M. (1988). Wild boar social groupings and their seasonal changes in the Camargue, southern France. *Zeitskrift fur Säugetierkunde*, 53, pp 22-30.

Delcroix, I., Mauget, R. & Signoret, J.P. (1990). Existence of synchronization of reproduction at the level of the social group of the European wild boar (*Sus scrofa*). *Journal of reproduction and fertility*, 89, pp 613-7.

Ebbert, W. & Bostedt, H. (1993). Cystic degeneration in porcine ovaries – First communication: Morphology of cystic ovaries, interpretation of the results. *Reproduction in Domestic Animals*, 28, pp 441 – 450.

Einarsson, S. & Gustafsson, B. (1970). Developmental abnormalities of female sexual organs in swine. *Acta Veterinaria Scandinavica*, 11, pp 427-442.

Einarsson, S., Brandt, Y., Lundeheim, N. & Madej, A. (2008). Stress and its influence on reproduction in pigs, a review. *Acta Veterinaria Scandinavica*, 50. Doi:10.1186/1751-0147-50-48.

Eliasson, L., Rydhmer, L., Einarsson, S. & Andersson, K. (1991). Relationship between puberty and production traits in the gilt: 1. Age at puberty. *Animal Reproduction Science*, 25, 143-154. Doi:10.1016/0378-4320(91)90039-3

Emanuelsson, U. & Liljegren, R. (2016). Landskapet, människan och djuret. In: Danell, K., Bergström, R., Mattsson, L. & Sörlin, S. (Eds) *"Jaktens historia" vilt - människa – samhälle*. pp 21-33. China: Liber AB. ISBN 978-91-47-11294-4.

Engblom, L., Lundeheim, N., Dalin, A.-M. & Andersson, K. (2007). Sow removal in Swedish commercial herds. *Livestock Science*, 106, pp 76-86.

European Commission. 2017. *African Swine fever – latest developments* [online]. Available from: http://ec.europa.eu/food/animals/animal-diseases/control-measures/asf/what-is-asf_en. Accessed 2017-04-19.

Evans, A.C.O. & O'Doherty, J.V. (2001). Endocrine changes and management factors affecting puberty in gilts. *Livestock Production Science*, 68, pp 1-12.

Fernández-Llario, P. & Mateos-Quesada, P. (1998). Body size and reproductive parameters in the wild boar (*Sus scrofa*). *Acta Theriologica*, 43, pp 439-444.

Fernández-Llario, P. & Carranza, J. (2000). Reproductive performance of the wild boar in a Mediterranean ecosystem under drought conditions. *Ethology Ecology and Evolution*, 12, pp 335-343. Doi:10.1080/08927014.2000.9522791.

Fonseca, C., Santos, P., Monzón, A., Bento, P., Alves da Silva, A., Alves, J., Silvério, A., Soares A. & Petrucci-Fonseca, F. (2004). Reproduction in the wild boar (*Sus scrofa* Linnaeus, 1758) populations of Portugal. *Galemys*, 16, pp 53-65.

Foxcroft, G.R., Dixon, W.T., Dyck, M.K., Novak, S., Harding, J.C.S. & Almeida, F.C.R.L. (2009). Prenatal programming of postnatal development in the pig. *Society for Reproduction and Fertility*, 66, pp 213- 231.

Frauendorf, M., Gethöffer, F., Siebert, U. & Keuling, O. (2016).The influence of environmental and physiological factors on the litter size of wild boar (*Sus scrofa*) in an agriculture dominated area in Germany. *Science of the Total Environment*, 541, pp 877–882. Doi:10.1016/j.scitotenv.2015.09.128.

Frichot, E. & François, O. (2015). LEA: an R package for landscape and ecological association studies. *Methods in Ecology and Evolution*, 6, pp 925-929. Doi:10.1111/2041-210X.12382.

Gaillard, J.M. & Jullien, J.M. (1993). Body weight effect on reproduction of young wild boar (*Sus scrofa*) females: a comparative analysis. *Folia Zoologica*, 42, pp 204-12.

Geisser, H. & Reyer, H.U. (2005). The influence of food and temperature on population density of wild boar *Sus scrofa* in the Thurgau (Switzerland). *Journal of Zoology*, 267, pp 89 – 96.

Gethöffer, F., Sodeikat, G. & Pohlmeyer, K. (2007). Reproductive parameters of wild boar (*Sus scrofa*) in three different parts of Germany. *European Journal of Wildlife Research*, 53, pp 287–97.

Gongora, J., Peltoniemi, O.A.T., Tammen, I., Raadsma, H. & Moran, C. (2003). Analyses of possible domestic pig contribution in two populations of Finnish farmed wild boar. *Acta Agriculturae Scandinavica, Section A – Animal Science*, 53, pp 161-165. Doi:10.1080/09064710310010602.

Gortázar, C., Ferroglio, E., Höfle, U., Frölich, K. & Vicente, J. (2007). Diseases shared between wildlife and lifestock: a European perspective. *European Journal of Wildlife Research*, 53, pp 241-256.

Hausman, G.J., Barb, C.R. & Lents C.A. (2012). Leptin and reproductive function. *Biochemie*, 94, pp 2075-2081. Doi:10.1016/j.biochi.2012.02.022.

Heinonen, M., Leppävuori, A. & Pyörälä, S. (1998). Evaluation of reproductive failure of female pigs based on slaughterhouse material and herd record survey. *Animal Reproduction Science*, 52(3), pp 235-44. Doi:10.1016/S0378-4320(98)00105-5.

Henry, V.G. (1968a). Length of estrous cycle and gestation in European wild hogs. *Journal of Wildlife Management*, 32, pp 406-408. Doi:10.2307/3798986.

Henry, V.G. (1968b). Fetal development in European wild hogs. *Journal of Wildlife Management*, 32, pp 966-970. Doi:10.2307/3799577.

Herrero, J., García-Serrano, A., Couto, S., Ortuño, V.M. & García-González, R. (2006). Diet of wild boar *Sus scrofa* L. and crop damage in an intensive agroecosystem. *European Journal of Wildlife Research*, 52, pp 245–250. Doi:10.1007/s10344-006-0045-3.

Hughes, PE. (1982). Factors affecting the natural attainment of puberty in the gilt. In: Cole, D.J.A. & Foxcroft, G.R. (Eds) *Control of pig reproduction*. pp 117-138. London: Butterworths.

Hultén, F., Zang, B.R., Forsberg, M. & Dalin, A.-M. (1995). Applying a progesterone assay to faecal samples collected from sows during the oetrous cycle. *Reproduction in Domestic Animals*, 30, pp 101-105. Doi:10.1111/j.1439-0531.1995.tb00613.x.

Hutchens, L.K., Hintz, R.L. & Johnson, R.K. (1982). Breed comparison for age and weight at puberty in gilts. *Journal of Animal Science*, 55, pp 60-66.

Hälli, O., Ala-Kurikka, E., Nokireki, T., Skrzypczak, T., Raunio-Saarnisto, M., Peltoniemi, O.A.T. & Heinonen, M. (2012). Prevalence of and risk factors associated with viral and bacterial pathogens in farmed European wild boar. *The Veterinary Journal*, 194, pp 98-101.

Jaktförordning, 1987:905 [online]. Available from: https://www.notisum.se/rnp/sls/lag/19870905.htm#B1. Accessed 2 Feb, 2017.

Ježek, M., Štípek, K., Kušta, T., Červený, J. & Vícha, J. (2011). Reproductive and morphometric characteristics of wild boar (*Sus scrofa*) in the Czech Republic. *Journal of Forest Science*, 57, pp 285-292.

Kaminski, G., Brandt, S., Baubet, E. & Baudoin, C. (2005). Life-history patterns in female wild boar (*Sus scrofa*): mother-daughter postweaning associations. *Canadian Journal of Zoology*, 83, pp 474-480. Doi:10.1139/Z05-019.

Kjellsson, M. (2017). Ny toppnotering i antalet fällda vildsvin. *Svensk Jakt*, 8, pp 48-49.

Langendijk, P. & Peltoniemi, O. (2013). How does nutrition influence luteal function and early embryo survival? In: Rodrigues-Martinez, H., Soede, N.M. & Flowers W.L. (Eds) *Control of pig reproduction IX*. pp 145-158. Packington, UK: Context products.

Lemel J. (1999). *Populationstillväxt, dynamik och spridning hos vildsvinet, Sus scrofa, i mellersta Sverige. Slutrapport*. Uppsala: Forskningsavdelningen, Svenska Jägareförbundet.

Lemel, J., Truvé, J. & Söderberg, B. (2003). Variation in ranging and activity behaviour of European wild boar *Sus scrofa* in Sweden. *Wildlife Biology*, 9, pp 29-36.

Love, R.J., G, Evans. & Kluoiec, C. 1993. Seasonal effects on fertility in gilts and sows. *Journal of reproduction and fertility*, 48, pp 191-206.

Lucia, Jr, T., Dial, G.D. & Marsh, W.E. (2000). Lifetime reproductive performance in female pigs having distinct reasons for removal. *Livestock Production Science*, 63, pp 213-222.

Länsstyrelsen Kalmar län. 2017. *Tillstånd bildförstärkare/lampa* [online]. Available from: http://www.lansstyrelsen.se/Kalmar/sv/djur-och-natur/jakt-och vilt/vildsvinsjakt/Pages/tillstand-bildf %c3 %b6rst %c3 %a4rkare-lampa.aspx. Accessed 2017-05-04.

Macchi, E., Starvaggi Cucuzza, A., Badino, P., Odore, R., Re, F., Bevilacqua, L. & Malfatti, A. (2010). Seasonality of reproduction in wild boar (*Sus scrofa*) were assessed by fecal and plasmatic steroids. *Theriogenology*, 73, pp 1230-1237.

Magnell, O. (2006). *Tracking wild boar and hunters, Osteology of wild boar in Mesolithic South Scandinavia*. Ph.D. Thesis, Department of Archaeology and Acient History, Lund University, Lund Sweden. ISBN 91-22-02141-8.

Malmsten, J., Söderquist, L., Thulin, C.G., Gavier Widén, D., Yon, L. & Dalin A.-M. (2014). Reproductive characteristics in female Swedish moose (Alces alces), with emphasis on puberty, timing of oestrus, and mating. *Acta Veterinaria Scandinavica*, 56, pp 1-10.

Markström, S. & Nyman, M. (2002). *Vildsvin*. Jägareförlaget. Kristianstads Boktryckeri AB, Kristianstad. ISBN 97-88660-44-3.

Massei, G., Genov, P.V. & Staines, B.W. (1996). Diet, food availability and reproduction of wild boar in a mediterranean coastal area. *Acta Theriologica*, 41, pp 307-320.

Mateusen, B., Maes, D.G.D., Van Soom, A., Lefebvre, D. & Nauwynck, H.J. (2007). Effect of porcine circovirus type 2 infection on embryos during early pregnancy. *Theriogenology*, 68, pp 896-901.

Matschke, G.H. (1967). Aging European wild hogs by dentition. *Journal of Wildlife Management*, 31, pp 109-113.

Mauget, R. (1972). Observations sur la reproduction du sanglier (*Sus scrofa* L.) a l'état sauvage. *Annales de biologie animale, biochimie, biophysique*, 12, pp 195-202.

Mauget, R. (1978). Seasonal reproductive activity of the European wild boar. Comparison with the domestic sow. In: Assenmacher, I. & Farner, D.S (Eds) *Environmental Endocrinology*. pp 79-80. Berlin, Heidelberg and New York: Springer Verlag. ISBN 978-3-642-66981-1.

Mauget R. 1982. Seasonality of reproduction in the wild boar. In: Cole, D.J.A. & Foxcroft, G.R. (Eds) *Control of Pig Reproduction*. pp 509-526. London: Butterworth Scientific.

Meng, X.J., Lindsay, D.S. & Sriranganathan, N. (2009). Wild boar as sources for infectious diseases in livestock and humans. *Philosophical Transactions of the Royal Society Biological Sciences,* 364, pp 2697-2707. Doi:10.1098/rstb.2009.0086.

Mengeling, W.L., Lager, K.M. & Vorwaldf, A.C. (2000). The effect of porcine parvovirus and porcine reproductive and respiratory syndrome virus on porcine reproductive performance. *Animal Reproduction Science*, 60–61, pp 199–210.

Miller, D.M. (1984). Cystic ovaries in swine. *The compendium on Continuing Education. Continuing Education Article*, 9, pp 31-35.

Moretti, M. (1995). Birth distribution, structure and dynamics of a hunted mountain population of wild boar (*Sus scrofa* L.), Ticino, Switzerland. *IBEX Journal of Mountain Ecology*, 3, pp 192-196.

Nationella Viltolycksrådet. (2017). *Statistik* [online]. Available from: http://www.viltolycka.se/statistik/. Accessed 2017-05-02.

Naturvårdsverket. (2010). *Nationell förvaltningsplan för vildsvin (Sus scrofa) - vägledning för regionala och lokala förvaltningsplaner*. ©Naturvårdsverket 200. ISBN 91-620- ISSN 0282-7298

Naturvårdsverket. (2017). *Jakt på vildsvin* [online]. Available from: http://www.naturvardsverket.se/Var-natur/Jakt/Jakt-pa-klovvilt/Jakt-pa-vildsvin/. Accessed 2017-05-04.

Olsson, U. (2002). *Generalized linear models – an applied approach*. Lund: Studentlitteratur;

Orlowska, L., Rembacz, W. & Florek, C. (2012). Carcass weight, condition and reproduction of wild boar harvested in north-western Poland. *Pest Management Science*, 69, pp 367-370. Doi:10.1002/ps.3355.

Pépin, D., Spitz, F., Janeau, G. & Valet, G. (1987). Dynamics of reproduction and development of weight in the wild boar (*Sus scrofa*) in South-West France. *Zeitskrift fur Säugetierkunde*. 52, pp 21-30.

Podgórski, T., Lusseau, D., Scandura, M., Sönnichsen, L. & Jędrzejewska, B. (2014). Long-lasting, kin-directed female interactions in a spatially structured wild boar social network. *PLoS ONE*, 9(6): e99875. Doi:10.1371/journal.pone.0099875.

Pozzi, P.S. & Alborali, G.L. (2012). Reproductive diseases in sows (*Sus scrofa domestica*): A review. *Israel Journal of Veterinary Medicine*, 67, pp 24-33.

R Core Team. (2013). *R: A Language and Environment for Statistical Computing*. R Foundation for Statistical Computing, Vienna. Available from: http://www.R-project.org /. ISBN 3-900051-07-0.

Risco, D., Llario, P.F., Velarde, R., García, W.L., Benítez, J.M., García, A., Bermejo, F., Cortés, M., Rey, J., de Mendoza, J.H. & Gómez, L. (2011). Outbreak of swine erysipelas in a semi-intensive wild boar farm in Spain. *Transboundary and Emerging Diseases*, 58, pp 445-450. Doi:10.1111/j.1865-1682.2011.01234.x.

Roberts, R.M., Xie, S. & Trout, W.E. (1993). Embryo-uterine interactions in pigs during week 2 of pregnancy. *Journal of Reproduction and Fertility. Supplement*, 48, pp 171-186.

Ruiz-Fons, F., Vicente, J., Vidal, D., Höfle, U., Villanúa, D., Gauss, C., Segalés, J., Almería, S., Montoro, V. & Gortázar, C. (2006). Sero-prevalence of six reproductive pathogens in European wild boar (*Sus scrofa*) from Spain: The effect on wild boar female reproductive performance. *Theriogenology*, 65, pp 731-743.

Ruiz-Fons, F. (2015). A review of the current status of relevant zoonotic pathogens in wild swine (*Sus scrofa*) populations: Changes modulating the risk of transmission to humans. *Transboundary and Emerging Diseases*. 64, pp 68-88. Doi:10.1111/tbed.12369.

Santos, P., Fernández-Llario, P., Fonseca, C., Monzón, A., Bento, P., Soares, A.M.V.M., et al. Habitat and reproductive phenology of wild boar (*Sus scrofa*) in the western Iberian Peninsula. *European Journal of Wildlife Research*, 52, pp 207-212.

SAS Institute Inc. (2011). *SAS/Stat User's Guide. Version 9*. Cary, N. C., SAS Institute Inc.

Servanty, S., Gaillard, J.M., Allaine, D., Brandt, D. & Baubet E. (2007). Litter size and foetal sex ratio adjustment in a highly polytocous species: the wild boar. *Behavioural Ecology*, 18, pp 427-432.

SMHI.se. 2016. *Vegetationperiod* [online]. Available from: http://www.smhi.se/kunskapsbanken/klimat/vegetationsperiod-1.6270. (Accessed June 30, 2016).

Stegeman, L.C. (1938). The European wild boar in the Cherokee National Forest, Tennessee. *Journal of Mammalogy*, 19, pp 279-290.

Stein, T.E., Dijkhuizen, A., D'Allaire, S. & Morris R.S. (1990). Sow culling and mortality in commercial swine breeding herds. *Preventive Veterinary Medicine*, 9, pp 85-94.

SVA. (2015). *Surveillance of infectious diseases in animals and humans in Sweden 2015*. National Veterinary Institute (SVA), Uppsala, Sweden. SVA: rapportserie 34 ISSN 1654-7098.

Tast, A., Hälli, O. & Peltoniemi, O.A. (2000). A practical method for non-surgically inserting intra arterial catheters in European wild boar (*Sus scrofa*). *Veterinary Research Communications*, 24, pp 25-30.

Thurfjell, H., Ball, J.P., Åhlén, P.A., Kornacher, P., Dettki, H. & Sjöberg, K. (2009). Habitat use and spatial patterns of wild boar *Sus scrofa*: agricultural fields and edges. *European Journal of Wildlife Research*, 55, pp 517-523.

Thurfjell, H., Spong, G. & Eriksson, G. (2014). Effects of weather, season, and daylight on female wild boar movement. *Acta Theriologica* , 59, pp 467–472.

Truvé, J. & Lemel, J. (2003). Timing and distance of natal dispersal for wild boar *Sus scrofa* in Sweden. *Wildlife Biology*, 9, pp 51-57.

Tummaruk, P., Kesdangsakonwut S. & Kunavongkrit A. (2009). Relationships among specific reasons for culling, reproductive data, and gross morphology of the genital tracts in gilts culled due to reproductive failure in Thailand. *Theriogenology*. 71, pp 369-375.

Tummaruk, P. & Kesdangsakonwut, S. (2014). Uterine size in replacement gilts associated with age, body weight, growth rate, and reproductive status. *Czech Journal of Animal Science*, 59, pp 511-518.

Vicente, J., Segalés, J., Höfle, U., Balasch, M., Plana-Durán, J., Domingo, M. & Gortázar, C. (2004). Epidemiological study on porcine circovirus type 2 (PCV-2) infection in the European wild boar (*Sus scrofa*). *Veterinary Research*, 35, pp 243-253. Doi:10.1051/vetres:2004008.

Vinsky, M.D., Murdoch, G.K., Dixon, W.T. & Dyck, M.K. (2007). Altered epigenetic variance in surviving litters from nutritionally restricted lactating primiparous sows. *Reproduction Fertility and Development*, 19, pp 430-435. Doi:10.1071/RD06082.

Welander, J. (2000). Spatial and temporal dynamics of wild boar (*Sus scrofa*) rooting in a mosaic landscape. *Journal of Zoology*, 252, pp 263 – 271.

Wikipedia.org. 2017. *Wild boar* [online], available from: https://en.wikipedia.org/wiki/Wild_boar. Accessed August 22 2017.

Wiklund, E. & Malmfors, G. (2014). *Viltkött som resurs*. Rapport 6635. © Naturvårdsverket Tryck: Arkitektkopia AB, Bromma. ISBN 978-91-620-6635-2 ISSN 0282-7298.

Wrathall, A.E. (1980). Ovarian disorders in the sow. Veterinary bulletin, 50, pp 253-272.

Wu, M.C., Hentzel, M.D. &, Dziuk, P.J. (1987). Relationships between uterine length and number of foetuses and prenatal mortality in pigs. *Journal of Animal Science*, 65, pp 762-770.

Yamamoto, K., Kijima, M., Takahashi, T., Yoshimura, H., Tani, O., Kojyou, T., Yamawaki, Y. & Tanimoto, T. (1999).Serovar, pathogenicity and antimicrobial susceptibility of *Erysipelothrix rhusiopathiae* isolates from farmed wild boars (*Sus scrofa*) affected with septicemic erysipelas in Japan. *Research in Veterinary Science*. 67, pp 301–303.

Zak, L.J., Cosgrove, J.R., Aherne, F.X. & Foxcroft, G.R. (1997). Pattern of feed intake and associated metabolic and endocrine changes differently affect postweaning fertility in primiparous lactating sows. *Journal of American Science*, 75, pp 208-216.

Zhang, C.-f., Song, C.-P., Chen, C.-M., Cui, S.-J. & Miao, L.-F. (2010). Reproductive failure in wild boar associated to porcine parvovirus infection and in vivo and in vitro characterization of the causal isolate. *Tropical Animal Health and Production*, 42, pp 1611-1613. Doi:10.1007/s11250-010-9644-9.

Østrup, E., Hyttel, P. & Østrup, O. (2011). Embryo-maternal communication: signalling before and during placentation in cattle and pig. *Reproduction, Fertility and Development*, 23(8), pp 964-975. Doi:org/10.1071/RD11140.

# Populärvetenskaplig sammanfattning

Vildsvin har varit en del av den svenska faunan under tusentals år men utrotades på 1600-talet på grund av högt jakttryck och domesticering. De vildsvin man idag ser i den svenska naturen stammar från ett fåtal djur som rymde från vilthägn under 1970- och 1980-talet. Det är svårt att veta exakt hur många vildsvin som finns i Sverige idag, men under de senaste åren har det skjutits cirka 100 000 djur per år. En uppskattad populationsstorlek på 200 000 djur kan därför inte anses vara överskattad. Om man jämför med andra klövviltsarter har vildsvinen en mycket hög reproduktionstakt (fortplantningsförmåga) och räknas till gruppen ”säsongsmässigt polyöstrala” djur. Detta betyder att deras brunst- och betäckningsperiod sker under en begränsad period. Vildsvinen räknas till gruppen ’short-day breeders’, vilket innebär att betäckningsperioden startar när dagarna blir kortare, d.v.s. under hösten. Enligt utländska studier varierar både reproduktionsmönster och reproduktionspotential (till exempel kullstorlek) mellan områden, länder och år. Faktorer som kan påverka vildsvinens reproduktion är bland annat födotillgång, hondjurets ålder och vikt, genetiskt ursprung, klimat och förekomst av smittämnen, till exempel virus och bakterier, i området. Syftet med denna studie var att erhålla ökad kunskap om vildsvins (hondjur) reproduktion under svenska förhållanden.

Mellan december 2012 och december 2015 organ och annat material (reproduktionsorgan, käkar/huvuden, blod/serum, magsäcksinnehåll, lever, njurar, öron) in från sammanlagt 617 vildsvin (hondjur) skjutna vid jakt på sju gårdar i fyra regioner (Blekinge, Skåne, Södermanland, Uppland). Alla gårdar bedriver kommersiell jakt och en del av förvaltningen innebär utfodring av vildsvinen mer eller mindre året om. Vildsvinen vägdes och djur med en kroppsvikt överstigande 30 kg inkluderades i studien. Vildsvinens ålder bedömdes utifrån tanduppsättning och tandväxlingsmönster. Reproduktionsorganen frystes in och undersöktes senare vid Sveriges lantbruksuniversitet (SLU). Livmodern och äggstockarna vägdes och mättes.

Alla strukturer i äggstockarna (folliklar, cykliska gulkroppar och dräktighetsgulkroppar) räknades och mättes. Utifrån strukturer i äggstockarna och livmoderns utseende och eventuellt innehåll bedömdes varje individs reproduktionsstadium. Vid förekomst räknades antalet embryon eller foster. Genom att mäta längden på dessa kunde dräktighetslängden (det vill säga tiden som djuret varit dräktigt) räknas ut, och i sin tur kunde brunst- och (om djuret hade fått leva) grisningsmånad beräknas. Förekomst av olika reproduktionsstörningar, såsom embryonal död, ombrunst, äggstockscystor och tecken på livmoderinflammation (endometrit) dokumenterades. Magsäcksinnehåll från 222 djur analyserades med avseende på innehållets ursprung; om det härrörde från fält, skogsmark eller från foderplats. Serum (blod) från 286 djur från Blekinge, Södermanland och Uppland analyserades med avseende på förekomst av antikroppar mot nio olika smittämnen, vilka direkt eller indirekt kan påverka reproduktionen.

Livmoderns vikt och längd varierade signifikant beroende på vilket reproduktionsstadie djuret befann sig i vid jakttillfället. Reproduktionsstörningar observerades hos ungefär 10 % av de undersökta vildsvinen. Dessa störningar skulle ha kunnat påverka djurens reproduktionsförmåga. Andelen könsmogna djur i åldern fem till femton månader ökade signifikant med stigande åldersklass och var årstidsberoende. Av alla djur inom detta ålderspann hade 16.6 % nått könsmognad. Detta var en lägre andel jämfört med vad tidigare europeiska studier visat med avseende på andelen könsmogna unga hondjur i områden med hög födotillgång.

Utifrån äggstocks- och livmoderfynd framkom att majoriteten av de undersökta djuren (alla åldersgrupper inkluderade) visade ett säsongsmässigt reproduktionsmönster. Det vill säga; anöstrala (inaktiva äggstockar) under sommaren, under hösten var äggstocksaktiviteten igång(brunst- och betäckningssäsong), och grisning under våren. Dock påträffades cykliska (djur med äggstocksaktivitet) och dräktiga djur under alla insamlingsmånader. Då månad för brunsttid beräknades kunde två brunsttoppar (i okt-nov och feb-mars) observeras. Att vissa djur inte följde det strikta säsongsmönster som karaktäriserar ’short day breeders’ kan exempelvis bero på att födotillgången var god (både som tillskottsfoder och föda från skog och odlad mark) och det genetiska ursprunget. Äggstocksfynd (tillbakabildad dräktighetsgulkropp) visade att vissa suggor hade förlorat sina kultingar under våren. Sådana djur kan i teorin hinna komma i brunst en gång till under våren, betäckas och föda en sommarkull och på så vis bidra till topp nummer två. Även gyltor som inte nått de fysiologiska förutsättningarna för att bli könsmogna under hösten kan ha kommit i brunst under våren med sommarkullar till följd. Medelkullstorleken, mätt som antalet foster eller embryon i livmodern i studien var 5.4 vilket är högt jämfört med vad som rapporterats från andra länder i Europa. Både reproduktionspotential och dräktighetsgrad ökade signifikant med suggornas ålder vilket är viktigt att ta hänsyn till i förvaltningen. Att vildsvinen grisar in under ’fel’ säsong i kombination med att suggor som är kultingförande är fredade från jakt i Sverige försvårar förvaltningen av djuren. För att förhindra att kultingförande suggor skjuts av misstag är ofta alla stora hondjur fredade vid jakt. Såsom studien visat innebär detta att de mest reproduktiva djuren, d.v.s. stora suggor, sparas vilket skulle kunna vara en bidragande orsak till den snabba ökningstakten av vildsvin i Sverige. Förekomsten av antikroppar mot vissa smittämnen (exempelvis porcint parvovirus och porcint circovirus typ 2) var hög eller mycket hög bland de undersökta djuren. Detta kan vara en effekt av höga tätheter samt aggregeringar av djur vid foderplatser vilket underlättar smittspridning. Dessa aspekter bör tas i beaktande generellt, och särskilt vid eventuell introduktion av en mer allvarlig smitta, exempelvis afrikansk svinpest.

Sammanfattningsvis kan sägas att reproduktionspotentialen hos de undersökta djuren var hög och påverkades av djurens vikt och ålder. Majoriteten av de undersökta djuren följde ett säsongsmässigt reproduktionsmönster, men avvikelser från detta förekom. Den generellt höga födotillgången och djurens genetiska bakgrund kan vara underliggande faktorer till dessa avvikelser. Täta populationer av vildsvin kan underlätta smittspridning och kan på så vis utgöra ett hot vid eventuell introduktion av allvarliga smittor.

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