UNIVERSITY OF THESSALY SCHOOL OF HEALTH SCIENCES FACULTY OF VETERINARY MEDICINE LABORATORY OF EPIDEMIOLOGY, BIOSTATISTICS AND ANIMAL HEALTH ECONOMICS

CHARACTERIZATION, QUANTIFICATION, IMPLICATIONS AND NUTRITIONAL MANAGEMENT OF SOW HOOF LESIONS: AN EPIDEMIOLOGIC DESCRIPTION AND ANALYSIS

A THESIS SUBMITTED FOR THE DEGREE OF DOCTOR OF PHILOSOPHY

MARINA LISGARA D.V.M

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EXAMINING COMMITTEE

Professor	L. Leontides	Supervisor
Professor	D. Maes	Member of Cons. Commitee
Assistant Professor	P. Kostoulas	Member of Cons. Commitee
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Assistant Professor	V. Papatsiros	Member of Exam. Commitee

SUMMARY

Chapter 1: Introduction

The introduction of the present thesis includes a review of current knowledge on sows' claw lesions, their frequency and associations with lameness and reproductive performance. At the end of the chapter the objectives of the thesis are presented.

Chapter 2: Hoof lesions and lameness in sows in three Greek swine herds

The purpose of the research presented in this chapter was, to characterize sows' hoof lesions, to estimate their frequency and severity and to investigate their associations with parity and lameness, in three Greek swine herds during the first 6 months of 2013. The results depict the baseline prevalence and severity of foot lesions before implementation of the directive for group housing in these herds.

The studied sows, which had been individually stalled during previous gestations, were examined for foot lesions upon entry into the lactation facilities. Lesions scored included heel hyperkeratinization, erosions or cracks, and toe and dew claw overgrowths. When exiting the farrowing facilities, the sows were observed while walking along an alley and their degree of lameness was scored. The proportion of sows with at least one lesion on any foot was very high and similar among herds, with 121 of 125 (96.8%), 123 of 125 (98.4%), and 377 of 386 (97.7%) sows affected in herds A, B, and C, respectively. The most frequent lesions were those located on the heel, and overgrown toes and dew claws. For

these sites, lesion severity increased with sow parity. The concurrent presence of lesions on more than one foot site, on the same or different feet or both, had a multiplicative effect on the likelihood of lameness. Under the conditions in the herds participating in this study, sow foot lesions are extremely common, with older sows more likely than younger sows to have lesions on the heel and overgrown toes and dew claws. The degree of lameness may be affected by a causal interface among foot lesions.

Chapter 3: Associations between claw lesions and reproductive performance of sows in three Greek herds

The aim of the study described in this chapter was to investigate the associations between the severity of sows' claw lesions and three of the most important reproductive indicators, the number of live-born and weaned piglets and the wean-to-first service interval, in three Greek farrow-to-finish herds.

All studied sows were individually housed during their previous gestations. Sows were examined for lesions, which were scored on a severity scale, on several anatomical sites of the claws, before farrowing. Data on the examined reproductive indicators were retrieved from productivity databases of the herds. Because scoring of lesions on several claw sites resulted in many correlated variables for each sow examined, we employed factor analysis to create a smaller set of uncorrelated variables (factors) which contained all the information in the original variables and produced the corresponding factor scores. The number of live-born and weaned piglets was associated with the produced factor scores in two multivariable linear regression models, whereas the possible association between the wean-to-first service interval and the factor scores was

modeled with the use of zero-inflated negative binomial regression. The number of live-born piglets was negatively associated with factor scores representing lesions on heel ($P \le 0.001$) and sole of front feet (P = 0.019). The number of weaned piglets was also negatively associated with factor scores representing lesions on heel (P = 0.003) of any foot, on sole of front feet (P = 0.001) and on white line, sole and wall of rear feet (P = 0.008), while the wean-to-first service interval was associated with factor scores representing lesions on heel of any foot (P = 0.02), on sole of front feet (P = 0.02) and of dew claw length of front feet (P = 0.009). Our results indicate that combinations of lesions on the dorsal and ventral part of the claws, negatively affected the reproduction parameters considered, emphasizing the importance of general improvement of feet health.

Chapter 4: Effect of diet supplementation with chelated zinc, copper and manganese on hoof lesions of loose housed sows

The objective of the research described in this chapter, was to investigate the effect of a diet supplemented with organic complexes of trace minerals (Zn, Cu, Mn), partly substituting their inorganic form, on hoof lesions of sows in three Greek swine herds.

A total of 518 sows were initially examined for hoof lesions and their respective severity was scored. For each hoof, the length of toes and dew claws were evaluated and five anatomical hoof sites, the heel, the sole, the white line, the wall and the coronary band, were examined for lesions. Subsequently, the same sows were re-scored after one or two gestations on diets supplemented with organic trace minerals, partly substituting their inorganic salt form (organic form of Zn 45 ppm, Cu 14 ppm and Mn 25 ppm of the total 125 ppm of Zn,

15 ppm of Cu and 40 ppm of Mn, respectively). The odds of the higher versus the lower lesion scores were significantly lower after than before the inclusion of the organic minerals in sows' diet, for each of the considered foot sites with the exception of the coronary band, with a distinct effect according to foot location. Specifically, on rear feet the improvement of hoof lesions was either smaller (for heel, sole and wall) than on front feet or not significant (for white line, toe and dew claw length). Additionally, for each foot site and herd examined, after the inclusion of the organic minerals, there were more sows with either the same or lower lesion score, with the exception of the toe and the dew claw length in one of the herds. Within the specific conditions in the three studied herds, the findings of this study highlight the role of chelated trace minerals in sows' hoof health, suggesting an applicable and rewarding intervention to prevent hoof lesions.

Chapter 5: General discussion and future perspectives

In this chapter the findings of the thesis are summarized and discussed with relevance to published research and the needs for future research activities.

ΠΑΝΕΜΙΣΤΗΜΙΟ ΘΕΣΣΑΛΙΑΣ ΣΧΟΛΗ ΕΠΙΣΤΗΜΩΝ ΥΓΕΙΑΣ ΤΜΗΜΑ ΚΤΗΝΙΑΤΡΙΚΗΣ ΕΡΓΑΣΤΗΡΙΟ ΕΠΙΔΗΜΙΟΛΟΓΙΑΣ, ΒΙΟΣΤΑΤΙΣΤΙΚΗΣ ΚΑΙ ΟΙΚΟΝΟΜΙΑΣ ΖΩΙΚΗΣ ΠΑΡΑΓΩΓΗΣ

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ΔΙΔΑΚΤΟΡΙΚΗ ΔΙΑΤΡΙΒΗ

ΜΑΡΙΝΑ ΛΙΣΓΑΡΑ ΚΤΗΝΙΑΤΡΟΣ

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ΕΠΤΑΜΕΛΗΣ ΕΞΕΤΑΣΤΙΚΗ ΕΠΙΤΡΟΠΗ

Καθηγητής	Λ. Λεοντίδης	Επιβλέπων
Καθηγητής	D. Maes	Μέλος Συμβ. Επιτροπής
Επίκουρος Καθηγητής	Π. Κωστούλας	Μέλος Συμβ. Επιτροπής
Καθηγητής	Γ. Αρσένος	Μέλος Εξετ. Επιτροπής
Καθηγητής	Χ. Μπάτζιος	Μέλος Εξετ. Επιτροπής
Επίκουρος Καθηγητής	Ε. Τζήκα	Μέλος Εξετ. Επιτροπής
Επίκουρος Καθηγητής	Β. Παπατσίρος	Μέλος Εξετ. Επιτροπής

ΠΕΡΙΛΗΨΗ

Κεφάλαιο 1: Εισαγωγή

Η εισαγωγή της παρούσας διατριβής περιλαμβάνει μια ανασκόπηση της υπάρχουσας γνώσης για τις αλλοιώσεις των χηλών των χοιρομητέρων, τη συχνότητα και τον συσχετισμό τους με τη χωλότητα και τις αναπαραγωγικές αποδόσεις. Στο τέλος του κεφαλαίου παρουσιάζονται οι σκοποί της διατριβής.

Κεφάλαιο 2: Αλλοιώσεις των χηλών και χωλότητα των χοιρομητέρων σε τρεις Ελληνικές εκτροφές

Ο σκοπός της μελέτης που παρουσιάζεται σε αυτό το κεφάλαιο ήταν, να χαρακτηριστούν οι αλλοιώσεις των χηλών των χοιρομητέρων, να υπολογιστεί η συχνότητα και η έντασής τους και να διερευνηθεί ο συσχετισμός τους με την ηλικία των χοιρομητέρων και τη χωλότητα, σε τρεις Ελληνικές εκτροφές, κατά τους πρώτους 6 μήνες του 2013. Τα αποτελέσματα απεικονίζουν τη συχνότητα και την ένταση των αλλοιώσεων των χηλών και της χωλότητας των χοιρομητέρων πριν την εφαρμογή του ομαδικού σταβλισμού στις εκτροφές.

Οι χοιρομητέρες που πήραν μέρος στη μελέτη και οι οποίες σταβλίζονταν ατομικά κατά τη διάρκεια των προηγούμενων κυήσεων, εξετάστηκαν για αλλοιώσεις στις χηλές κατά την είσοδό τους στους θαλάμους τοκετών και γαλουχίας. Οι αλλοιώσεις που αξιολογήθηκαν περιελάμβαναν την υπερκερατινοποίηση του βολβού της οπλής, διαβρώσεις ή ρωγμές, και υπερανάπτυξη των χηλών και επικουρικών χηλών. Ο έλεγχος της ύπαρξης χωλότητας και η βαθμολόγηση της έντασή της γίνονταν κατά την έξοδο των ζώων από τους θαλάμους τοκετών, επισκοπώντας τον τρόπο βάδισής των

χοιρομητέρων καθώς περπατούσαν κατά μήκος ενός διαδρόμου. Το ποσοστό των χοιρομητέρων με τουλάχιστον μια αλλοίωση σε οποιοδήποτε άκρο ήταν πολύ υψηλό και παρόμοιο μεταξύ των εκτροφών, με 121 από 125 (96.8%), 123 από 125 (98.4%), και 377 από 386 (97.7%) προσβεβλημένες χοιρομητέρες στις εκτροφές Α, Β, και Γ, αντίστοιχα. Οι πιο συχνές αλλοιώσεις ήταν εκείνες του βολβού της οπλής, και οι υπερμεγέθης χηλές και επικουρικές χηλές. Η ένταση αυτών των αλλοιώσεων αυξάνονταν με την πάροδο της ηλικίας των χοιρομητέρων (αριθμός τοκετών). Η ταυτόχρονη παρουσία αλλοιώσεων σε περισσότερα από ένα ανατομικά μέρη της χηλής, στο ίδιο ή διαφορετικό άκρο ή και τα δύο, είχαν πολλαπλασιαστικό αποτέλεσμα στην πιθανότητα εμφάνισης χωλότητας. Κάτω από τις συνθήκες των εκτροφών που έλαβαν μέρος στην παρούσα μελέτη, οι αλλοιώσεις των χηλών των χοιρομητέρων είναι εξαιρετικά συχνές, με τις μεγαλύτερες σε ηλικία χοιρομητέρες να είναι πιο πιθανό να εμφανίσουν αλλοιώσεις στο βολβό της οπλής και υπερμεγέθης χηλές και επικουρικές χηλές σε σχέση με τις νεαρότερες χοιρομητέρες. Ο βαθμός της χωλότητας μπορεί να επηρεαστεί από αιτιολογικές διασυνδέσεις μεταξύ αλλοιώσεων των χηλών.

Κεφάλαιο 3: Συσχετισμοί μεταξύ των αλλοιώσεων των χηλών και των αναπαραγωγικών αποδόσεων των χοιρομητέρων σε τρεις Ελληνικές εκτροφές

Ο σκοπός της μελέτης που περιγράφεται σε αυτό το κεφάλαιο ήταν να διερευνηθούν οι συσχετισμοί μεταξύ της έντασης των αλλοιώσεων των χηλών των χοιρομητέρων και τριών από τους πιο σημαντικούς αναπαραγωγικούς δείκτες, τον αριθμό των ζωντανών γεννηθέντων και απογαλακτισμένων χοιριδίων

και το μεσοδιάστημα από τον απογαλακτισμό μέχρι την πρώτη τεχνητή σπερματέγχυση, σε τρεις συμβατικές Ελληνικές εκτροφές.

Οι χοιρομητέρες που πήραν μέρος στη μελέτη σταβλίζονταν ατομικά κατά τη διάρκεια των προηγούμενων κυήσεών τους. Οι χοιρομητέρες ελέγχθηκαν για την ύπαρξη αλλοιώσεων, οι οποίες βαθμολογήθηκαν ανάλογα με τη βαρύτητά τους, σε διάφορα ανατομικά στοιχεία της χηλής, πριν τον τοκετό. Τα δεδομένα των υπό εξέταση αναπαραγωγικών δεικτών προήλθαν από τα αρχεία της κάθε εκτροφής. Επειδή η βαθμολόγηση των αλλοιώσεων στα διάφορα ανατομικά μέρη των χηλών οδήγησε στη δημιουργία πολλών συσχετιζόμενων μεταξύ τους μεταβλητών για κάθε χοιρομητέρα που εξετάστηκε, χρησιμοποιήσαμε την παραγοντική ανάλυση για να δημιουργήσουμε ένα μικρότερο σύνολο ανεξάρτητων μεταβλητών (παραγόντων - factors), το οποίο περιείχε όλες τις πληροφορίες των αρχικών μεταβλητών. Στη συνέχεια υπολογίσαμε τους συντελεστές των παραγόντων. Ο αριθμός των ζωντανών γεννηθέντων και απογαλακτισμένων χοιριδίων συσχετίστηκε με τους συντελεστές των παραγόντων σε δυο πολυπαραγοντικά μοντέλα γραμμικής παλινδρόμησης, ενώ οι πιθανοί συσχετισμοί μεταξύ του διαστήματος απογαλακτισμού-οίστρου και τους συντελεστές των παραγόντων ελέγχθηκαν με τη χρήση ενός αρνητικού διωνυμικού μοντέλου μηδενικού-πληθωρισμού (zero-inflated negative binomial model). Ο αριθμός των γεννηθέντων ζωντανών χοιριδίων συσχετίστηκε αρνητικά με συντελεστές των παραγόντων που αντιπροσώπευαν αλλοιώσεις στο βολβό της οπλής (P ≤ 0.001) και στη σόλα των προσθίων άκρων (P = 0.019). Ο αριθμός των απογαλακτισμένων χοιριδίων συσχετίστηκε επίσης αρνητικά με συντελεστές παραγόντων που αντιπροσώπευαν αλλοιώσεις στο βολβό της

οπλής (P = 0.003), στη σόλα των προσθίων άκρων (P = 0.001) και στη λευκή γραμμή, στη σόλα και στο τοίχωμα των οπισθίων άκρων (P = 0.008), ενώ το μεσοδιάστημα απογαλακτισμού μέχρι την πρώτη σπερματέγχυση συσχετίστηκε με αλλοιώσεις στο βολβό της οπλής (P = 0.02), στη σόλα των προσθίων άκρων (P = 0.02) και στο μήκος των επικουρικών χηλών στα πρόσθια άκρα (P = 0.009). Τα αποτελέσματά μας έδειξαν ότι ο συνδυασμός διαφόρων αλλοιώσεων στο ραχιαίο και κοιλιακό τμήμα των χηλών, επηρεάζει αρνητικά τις αναπαραγωγικές παραμέτρους, τονίζοντας τη σημασία της υγείας των ποδιών και την ανάγκη γενικής βελτίωσής τους.

Κεφάλαιο 4: Επίδραση της προσθήκης χηλικού ψευδαργύρου, χαλκού και μαγγανίου στις αλλοιώσεις των χηλών χοιρομητέρων που σταβλίζονται ομαδικά

Το αντικείμενο της μελέτης που περιγράφεται σε αυτό το κεφάλαιο ήταν, να διερευνηθεί η επίδραση μιας δίαιτας εμπλουτισμένης με οργανικά σύμπλοκα ιχνοστοιχείων (Zn, Cu, Mn), υποκαθιστώντας μερικώς την ανόργανη μορφή τους, στις αλλοιώσεις των χηλών των χοιρομητέρων τριών Ελληνικών εκτροφών.

Συνολικά 518 χοιρομητέρες εξετάστηκαν για αλλοιώσεις στις χηλές και η έντασή τους βαθμολογήθηκε. Για κάθε χηλή, το μήκος των χηλών και επικουρικών χηλών αξιολογήθηκε και πέντε ανατομικά στοιχεία, ο βολβός της οπλής, η σόλα, η λευκή γραμμή, το τοίχωμα και η στεφάνη, εξετάστηκαν για αλλοιώσεις. Στη συνέχεια, οι ίδιες χοιρομητέρες επανεξετάστηκαν μετά από έναν ή δυο τοκετούς σε δίαιτα εμπλουτισμένη με οργανικά ιχνοστοιχεία, υποκαθιστώντας μερικώς την ανόργανη μορφή τους (οργανική μορφή Zn 45 ppm, Cu 14 ppm και Mn 25 ppm από τη συνολική ποσότητα 125 ppm του Zn,

15 ppm του Cu και 40 ppm του Mn, αντίστοιχα). Η πιθανότητα για μεγαλύτερης έντασης αλλοιώσεις σε σχέση με μικρότερης έντασης ήταν σημαντικά μικρότερη μετά σε σχέση με πριν την προσθήκη των οργανικών ιχνοστοιχείων στη δίαιτα των χοιρομητέρων, για το καθένα από τα ανατομικά στοιχεία που εξετάστηκαν με εξαίρεση τη στεφάνη, και με διακριτή επίδραση ανάλογα με την εντόπισή τους. Συγκεκριμένα, στα οπίσθια άκρα η βελτίωση των αλλοιώσεων των χηλών ήταν είτε μικρότερη (για το βολβό της οπλής, τη σόλα και το τοίχωμα) σε σχέση με τα πρόσθια άκρα ή δεν ήταν σημαντική (για τη λευκή γραμμή, το μήκος των χηλών και επικουρικών χηλών). Επιπλέον, για κάθε ανατομικό στοιχείο και εκτροφή που εξετάστηκε, μετά την προσθήκη των οργανικών ιχνοστοιχείων, οι χοιρομητέρες με την ίδια ή μικρότερη ένταση αλλοιώσεων ήταν περισσότερες, με εξαίρεση το μήκος των χηλών και επικουρικών χηλών στη μια από τις εκτροφές. Κάτω από τις συγκεκριμένες συνθήκες των τριών εκτροφών που μελετήθηκαν, τα ευρήματα της παρούσας μελέτης επισημαίνουν τον ρόλο των χηλικών ιχνοστοιχείων στην υγεία των χηλών των χοιρομητέρων, προτείνοντας μια εύκολα εφαρμόσιμη και αποδοτική παρέμβαση για την πρόληψη των αλλοιώσεων των χηλών.

Κεφάλαιο 5: Γενική συζήτηση και προοπτικές για το μέλλον

Στο κεφάλαιο αυτό συνοψίζονται και συζητούνται τα συμπεράσματα της διατριβής σε σχέση με την υπάρχουσα βιβλιογραφία και τις ανάγκες για μελλοντικές ερευνητικές δραστηριότητες.

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Index

SUMMARY	3
ΠΕΡΙΛΗΨΗ	
Acknowledgments	
Index	
Chapter 1	
1. Introduction	
1.1 The pig's foot	
1.2. Hoof lesions	
1.3. Factors associated with claw lesions	
1.3.1. Claw conformation	25
1.3.2. Flooring and housing	27
1.3.3. Nutrition	
1.4. Foot lesions and reproductive performance	33
1.5. Background and Objectives	
1.6. References	
Chapter 2	51
2.1. Summary	53
2.2. Introduction	54
2.3. Materials and methods	55
2.3.1. Study population	55
2.3.2. Study design	56
2.3.3. Statistical analysis	61
2.4. Results	65
2.4.1. Foot lesions	65
2.4.2. Association of lesions with parity	69
2.4.3. Association of lesions with lameness	71
2.5. Discussion	74
2.6. Conclusions	78
2.7. References	78
Chapter 3	84
3.1. Summary	86
3.2. Introduction	87

3.3. Materials and methods	88
3.3.1. Study population	88
3.3.2. Study design	88
3.3.3. Statistical analysis	90
3.4. Results	94
3.5. Discussion	98
3.6. Conclusions	101
3.7. References	101
Chapter 4	107
4.1. Summary	109
4.2. Introduction	110
4.3. Materials and Methods	113
4.3.1. Study population	113
4.3.2. Study design	113
4.3.3. Statistical analysis	116
4.4. Results	117
4.5. Discussion	125
4.6. Conclusions	131
4.7. References	131
Chapter 5	140
5. General discussion and future perspectives	141
Appendix	146
I. A photographic documentation of the scoring system applied for the evaluation of lesions on seven sites of the sow's foot	147
II. List of publications from the present thesis	154

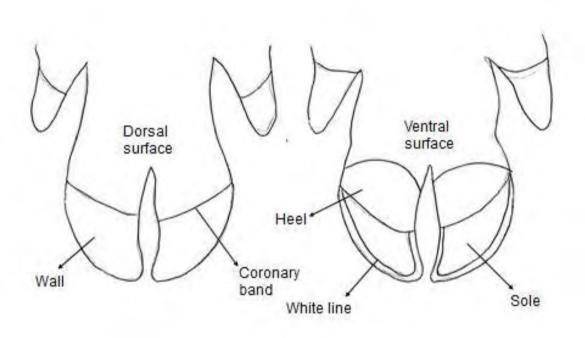
Chapter 1

1. Introduction

1.1 The pig's foot

The pig is tetradactylous having two large weight bearing digits, and two non weight bearing accessory digits or dewclaws (DCs); the first digit is absent in the pig. The third digit is medial and the fourth is lateral on each foot. The term foot refers to the entire region from the toe to the top of the DCs. Each digit includes the claw or hoof, which describes the keratinized horn tissue consisting of the hard wall, hard sole and soft heel. Each claw covers the third phalanx and the distal parts of the second phalanx, i.e. the distal phalanges, together with their horn capsule, form the claws and dew claws, respectively (Nalon et al., 2013). The DCs are caudal to the larger digits and have a full complement of bones (phalanges). Four distinct types of horn make up the different anatomical sites of the claws. The hard outer covering of the claw is called the hoof wall and a slightly softer region underneath the hoof is called the sole (Hepworth et al., 2004). The junction between the sole and the wall constitutes the white line, which allows the hoof to be flexible as the animal moves (Hepworth et al., 2004). Posterior to the sole on the caudal end of the toe lies the heel, which consists of soft keratinized tissue. The point where the hoof meets the hairy skin on the animal's foot is called the coronary band. The anatomical sites of the hoof are depicted in Figure 1.

Figure 1. The anatomical sites on the dorsal and the ventral surface of the sow's hoof.



A cross-section of the claw consists of bone of the phalanx, subcutus, corium or dermis, vascular basement membrane, and epidermis. The corium located just below the epidermis forms the supportive connective tissue layer for the epidermis, containing blood vessels and nerves. From this layer, nutrients and hormones are provided to the stratum basale for the production of the epidermal cells. All distal layers of the epidermis are derived from these cells by a process of proliferation and differentiation. The corium is critical for keratinized horn formation. Keratins are produced by a complex process of differentiation (termed "keratinization") of the epidermal cells. The hallmarks of keratinization

are formation and biochemical binding of keratin proteins, and synthesis and exocytosis of intracellular cementing substance (Tomlinson et al., 2004).

The corium is covered by the densely arranged cells of the stratum basale, which are pushed into the next layer and enter the process of differentiation to make up the stratum spinosum. Toward the end of differentiation, basophilic dense keratohyalin granules accumulate in the cells, making up the stratum granulosum. This layer forms the border of cornification during which the cells die (that is cornify) and turn into horn cells, establishing the stratum corneum. The previously outlined process is dependent on an appropriate supply of nutrients, including vitamins, minerals and trace elements. They are essential for the integrity of the hoof horn. Anything decreasing the supply of nutrients to keratinizing epidermal cells can lead to inferior horn quality and increase its susceptibility to physical, chemical, or microbial damage from the environment (Tomlinson et al., 2004), which may result in clinical lesions of the claws and associated lameness.

1.2. Hoof lesions

Hoof lesions may involve the heel, sole, white line, wall and DCs. The characteristics of pigs' weight distribution and gait, together with the biomechanical characteristics of each specific type of horn, make the different anatomical sites more or less susceptible to the development of cracks (heel, sole-heel junction, white line, claw wall), erosions (heel) and overgrowth (toes, DCs) (Nalon et al., 2013). Gjein and Larssen (1995a) were the first to describe a claw lesion classification system, which included the evaluation of six main types

of lesions, namely cracks in the sidewall, white line and heel-sole junction separation, heel overgrowth and lesions in the heel and toe cracks, on a severity scale with five categories. In more recently applied scoring systems, claw and dew claw overgrowth and/or lesions on the skin above the coronary band were also evaluated (Foot First Team, 2010), and lesion severity scales with three (Bradley et al., 2007; Foot First Team, 2010) or four categories were used (Deen and Winders, 2008; Pluym et al., 2011).

Heel lesions start as bruising seen as dark discoloured areas under the volar surface. This is followed by erosion, which appears as a rough "ragged" lesion with underlying haemorrhage. Ulceration of the erosion may ensue. Continuous trauma may result in hyperkeratinisation, excessive granulation, and necrosis often extending in the interdigital cleft (Penny et al., 1963). Sole lesions are also initially seen as bruising with dark brown to red areas of hemorrhage under the surface, most frequently observed along the heel and sole junction. Hyperkeratinization of the sole can be a sequel to erosion and ulceration. White line lesions of the claw start as hard tissue wear and separation between the hard and soft tissue of the hoof wall and sole and heel or both (Bradley et al, 2008). These lesions may result in extensive separation. The separation may be filled with dirt, feces and other foreign material often resulting in painful infection and inflammation of the corium. Cracks of the wall can be vertical starting at the white line or starting at the coronary band or horizontal (Penny et al., 1963). Coronary band abscesses are most likely an infection of the laminae. The area above the coronary band becomes swollen producing a purulent discharge. DC lesions, which are common in sows, may involve loss of the horn with

hemorrhage and infection of the corium or various levels of lengthening of the digit.

The reported frequency of occurrence of hoof lesions of sows in modern swine herds is very high, usually exceeding 90% of the studied sow populations (Anil et al., 2007; Enokida et al., 2011; Pluym et al., 2011). More than 50% of the examined sows had moderate or severe lesions (Gjein and Larssen, 1995a; Enokida et al., 2011; Fitzgerald et al., 2012). Hoof abnormalities occurred more frequently and severely in older sows (Dewey et al., 1993; Pluym et al., 2011; Fitzgerald et al., 2012) although a reverse effect has also been reported (Anil et al., 2007), probably due to the differential culling rate of affected sows.

Claw lesions were considered an important underlying cause of locomotor disorders (Anil et al., 2007), because specific types of claw lesions were associated with an increased risk of lameness (Gjein and Larssen, 1995b; Anil et al., 2007). Claw lesions were found to be correlated with inflammation of the corium in lame sows (Cameron, 2012). Although the pathology and etiology of inflammation may not be the same for each lesion, because of the large numbers of nerves associated with the corium, inflammation is very likely to result in pain and lameness. Lameness is the clinical manifestation of a series of locomotor disorders that are frequently encountered in many production animal species, including breeding sows (Nalon et al., 2013). It is characterised by alterations in the normal gait and posture and by a reduced mobility due to pain or discomfort (Bourne, 2011). The simplest locomotion scoring methods rely on the ability of trained observers to visually identify various degrees of deviation from a "normal" gait, while sow is walking on solid floor (Nalon et al., 2013). The most typical

signs of lameness include, reluctance to move, reduced walking speed, shorter or uneven stride length, swaying from side to side and in cases of severe lameness vocalisations (squealing) (Karlen et al., 2007; Okholm Nielsen, 2011; Grégoire et al., 2013). A hunched posture (arched back) can sometimes be observed (Grégoire et al., 2013) and in severe cases, there is minimal or no weight bearing on the affected limb(s) or the animal refuses to move (Main et al., 2000). Other postural or gait abnormalities such as uneven weight bearing, asymmetric stance and "goose stepping" can also be observed (Jørgensen, 2000; Kirk et al., 2008). Nonetheless, visual gait scoring in sows can be challenging because this species tends to hide overt signs of lameness (Okholm Nielsen, 2011). Head bobbing may be present but, in contrast to cows, is generally difficult to identify due to the short necks of pigs, which limits the vertical movement of the head (Main et al., 2000). Scores of lameness are commonly assigned on ordinal scales. Most of the scoring systems described in the literature comprise four (Karlen et al., 2007; Zinpro Foot First, Swine locomotion scoring system) or five (Geverink et al., 2006; Mustonen et al., 2011; Grégoire et al., 2013) categories, whereas the Welfare Quality Protocol® (2009) includes three categories for the evaluation of the locomotor ability of the sows.

Lameness may result from various pathologies or injuries to the foot, the bones or the joints (Wells, 1984; Dewey et al., 1993) such us osteochondrosis, arthrosis or arthritis, broken legs and feet injuries (D'Allaire and Drolet, 2006). Approximately 5% to 20% of lameness cases in sows were attributable to feet lesions (Dewey et al., 1993; Kirk et al., 2005). Location (Anil et al., 2007; 2008) and severity (Gjein and Larssen, 1995b; Plyum et al., 2011) of the lesions might

determine whether a sow shows overt lameness or not. Anil et al. (2007) reported that sows with white line lesions were more likely to be lame, while other hoof lesions were not associated with lameness. Heel, sole and white line lesions affected the gait, footprints or stepping of sows, in a study that kinematics and footprint analysis were used to evaluate sows' locomotor ability (Grégorie et al., 2013). Moreover, Hulten (1995) and Jørgensen (2000) reported a correlation between hoof overgrowth and sow locomotor disorders. In a more recent study, lame sows had significantly higher dew claw length compared to non lame sows (Pluym et al., 2011). In contrast to these findings, Kroneman et al. (1993) and Bradley et al. (2009) found no relationship between hoof lesions and lameness.

Lameness is considered an important animal-based welfare indicator (Welfare Quality Protocol®, 2009), since it can affect the general health of the sow by causing stress and pain (Heinonen et al., 2013). High levels of corticosteroids in blood can reduce proliferation of lymphocytes and decrease antibody production and therefore impair the ability to resist infection (Kelley, 1980). Moreover, the daily feed intake of the lame sow is reduced due to loss of appetite and her locomotor disability (Fitzgerald et al., 2012). Therefore, the sow becomes more susceptible to other diseases due to her decreased immunological response (Heinonen et al., 2013). Lameness also affects sow longevity, since it is the most commonly reported cause after reproductive reasons for premature culling of breeding sows (Anil et al., 2005; Kirk et al., 2005; Engblom et al., 2008). Acutely lame sows are removed immediately from the herd, and chronic, less severe lameness can affect the performance of sows and thus indirectly lead to sow removals (Anil et al., 2009). On a herd level, this

can be seen as decrease in reproductive results and increase in culling or euthanasia of sows (Heinonen et al., 2013).

1.3. Factors associated with claw lesions

1.3.1. Claw conformation

Penny et al. (1963) measured claw size and found that the average ratio of length of lateral claw to medial claw was 1.11:1 and width 1.13:1. They found significantly more lesions on the lateral claw especially on the hind feet. They believed that the difference in claw size was most likely hereditary and that the smaller medial claw was somewhat protected by the larger lateral claw. Therefore, the medial claws take less weight and are less frequently traumatized than the lateral claws and sows with smaller inner hooves may have greater chance to develop claw lesions due to their uneven weight distribution (Kornegay et al., 1990; Kroneman et al., 1993; Enokida et al., 2011). The severity of hoof lesions was reported higher on rear than front feet and on lateral than medial claws (Gjein and Larssen, 1995a; Kirk et al., 2005; Anil et al., 2007). Lateral claws tend to be larger than medial claws with the discrepancy in size being more pronounced on rear feet than on front feet and increasing as pigs' age (Penny et al., 1963; Bradley et a., 2007; Amstel and Doherty, 2010). Inequality of the size of the claws and varying tissue strength between medial and lateral claws contribute to the difference in susceptibility (Webb, 1984; Tubbs, 1988; Kroneman et al., 1993; Gjein and Larssen, 1995a; Pluym et al., 2013a). Hence, hoof lesions may be more common on lateral claws also because of a greater

weight-bearing surface than medial claws (Tubbs, 1988). Interestingly, more than 75% of the weight of the pigs is born by the lateral digit and 80% of the injuries affect these digits (Webb, 1984). Moreover, junctions between hard and soft areas of the horn may be more susceptible to injury than other areas of the hoof horn (Kroneman et al., 1993). Maximum weight is born by the heel bulb, followed by the junction between the heel bulb and abaxial hoof wall of the lateral digit. Amstel et al. (2009) reported that the front feet have better weight-bearing stability than the rear feet, with the rear medial claw as the least stable. It was suggested that this may result in more weight being transferred to the rear lateral claw, which may predispose this claw to the higher observed incidence of lesions.

The disparity in claw size in piglets would suggest it is hereditary (Penny et al., 1963). Fan et al. (2009) identified possible genetic markers associated with uneven claw size. Genetic selection of replacement gilts with even-sized toes may help to control the development of claw lesions (Pluym et al., 2013a).

Apparently, because of its relevant genetic background and the impact on sow culling, hoof growth could be an appealing selection criterion for prevention of claw lesions (Pluym et al., 2013a), especially hoof overgrowth. Lastly, during gilt selection, examining their stance, gait and hoof status is of great importance for their future productivity and longevity (Stalder, 2010). Strong and even legs, straight back, uniform hooves and normal gait are crucial for a sow when starting her reproductive life (Stalder, 2010). Buck-kneed front legs or post-legged position of rear legs may predispose to foot pad abrasions, swelling of the feet and joints, and lameness (Stalder, 2010).

1.3.2. Flooring and housing

The impact of housing and flooring on claw lesion development has been the subject of intensive research in swine. A higher incidence of claw lesions in group compared to individually housed sows has been previously reported (Anil et al., 2007). However, the EU Directive 2001/88/EC, implemented since January 2013 in all 25 member states, requires that sows and gilts be kept in groups during a period starting 4 weeks after service and until 1 week before the expected time of farrowing. Deep straw bedding, which has been reported to reduce the incidence and severity of claw lesions (Gjein and Larssen, 1995c; Ehlorsson et al., 2002; Kilbride et al., 2010), could be used in loose housing of pregnant sows. However, it does not guarantee full protection against hoof lesions. The lack of natural wear of the horn on solid floors with deep bedding might increase the risk of toe overgrowth and erosions, especially when straw is wet and grubby (Kilbride et al., 2009; Pluym et al., 2013a). Moreover, the use of group-housing on deep straw bedding can be expensive, the manure system on many farms is incapable of handling large amounts of litter (Pluym et al., 2013a) and is not applicable in temperate climates and in partially or fully slatted flooring.

Although slatted concrete floors have been accused for higher incidence of lameness compared to solid concrete floors, they are most often used for group-housing of gestating sows, since they do not require manual removal of soiled bedding and are less costly and labor intensive (Pluym et al., 2013a). In this type of flooring the quality and hygiene of the floor are of major importance to control the incidence of hoof lesions. Slip-resistance, abrasiveness and void ratio (i.e. the ratio of area of holes per unit area of floor) are the main characteristics

contributing to the injury potential of a fully or partially slatted floor (McKee and Dumelow, 1995). Regarding the void ratio, the Council Directive of 2001(2001/88/CE) imposed a maximum slot width of 2.0 cm, whereas a previous study suggested slot width of 1.3-1.6 cm for minimizing the incidence of claw lesions in sows (Tubbs, 1988). However, due to floor wearing, the gap width of the slot increases and crumbled slat edges are formed in old floors. Use of plastic slats instead of concrete ones could be a solution, however, they are not recommended since they were not found to improve claw health in group-housed sows (Gjein and Larssen, 1995c; Olsson and Svendsen, 2002). In a recent study, rubber flooring appeared to improve heel overgrowth and erosion, heel-sole cracks, white line lesions and claw length, in group housed sows (Bos et al., 2016). Regarding floor hygiene, dry and clean floors decrease the risk of claw infections compared to wet and grubby ones (Gjein and Larssen, 1995b), which impair hoof strength and cause irritation of the soft heel tissue, leading to heel erosions and overgrowth (Webb, 1984; Kroneman et al., 1993; Gjein and Larssen, 1995a).

1.3.3. Nutrition

Despite the fact that housing and flooring conditions and managerial and genetic factors can affect the hoof health status (Kroneman et al., 1993), the role of specific nutrient intake is vital in developing the feet structure and integrity (Tomlinson et al., 2004). Keratinization of hoof epidermis is controlled and moderated by a variety of bioactive molecules and hormones. Minerals (Zn, Cu, Mn, Se, Ca) and vitamins (A, D, C, E and biotin) have a substantial contribution in production and preservation of healthy keratinized tissues (Mulling et al., 1999;

Van Riet et al., 2013). Except for biotin, however, very little peer-reviewed literature related to nutritional requirements for hoof growth in the pig is available and present knowledge is primarily based on hoof development and integrity researched in cattle and horses. Although the effect of biotin supplementation has been thoroughly investigated there is a controversy regarding the beneficial effect of biotin supplementation on the hooves of growing gilts or adult sows (Webb et al., 1984; Simmins and Brooks, 1988; Watkins et al., 1991). In more detail, early findings by Brooks et al. (1977) and Penny et al. (1980) demonstrated a decrease in number and in severity of feet lesions. Subsequent studies (Misir and Blair, 1986; Simmins and Brooks, 1988) also reported the benefit of biotin supplemented rations in relation to hoof lesions, hoof wall hardening, and foot pad resilience. The results showed that the response to biotin was dose and time dependent. In contrast, in a study by Watkins et al. (1991) the biotin supplemented ration did not affect the incidence and severity of foot lesions, although there was a tendency for lower lesion scores in the biotinsupplemented sows. Also Brooks and Simmins (1980) found no difference in horn strength as a result of biotin supplementation in induced biotin-deficient pigs. Kornegay (1986) reviewed evidence that biotin increases the hardness of the hoof wall but decreases the hardness of the heel bulb tissue, suggesting that biotin supplementation could decrease foot lesions on some foot sites and increase lesions on other foot sites. Additionally, the discrepancy in the efficiency of biotin supplementation seen in the literature most likely reflects the multifactorial etiology of foot lesions and the wide range of environmental, nutritional, and management practices used in the swine industry.

Zinc, copper and manganese were identified as key minerals in the processes of keratinization (Smart and Cymbaluk, 1997; Mulling et al., 1999; Mülling, 2000) since they have an instrumental role in the activation of enzymes with catalytic, structural and regulatory function (Cousins, 1996; Van Riet et al., 2013). They have functional implications in growth and development, reproduction and general health of the sow as well. Zinc supplementation in dairy cows has shown a reduction in cases of foot rot, heel cracks, interdigital dermatitis, and laminitis (Moore et al., 1989). Also in cattle, copper deficiency can result in heel cracks, foot rot, and sole abscesses. Manganese is essential for skeletal and tendon development, and joint and cartilage strength. Sows' mineral reserves decline over several reproductive cycles and depletion is exacerbated for hyper-prolific sows which are able to support larger litter growth rates (Mahan and Newton, 1995). Mineral absorption increases during lactation in response to the high nutrient demand for milk production, but when dietary mineral intake is insufficient, the sow will mobilize her body mineral reserves, particularly from the liver to meet the demand (Mahan and Newton, 1995). An inadequate mineral intake may affect hormonal secretion, enzyme activity, muscle function, bone mineral content, and other body mineral functions (Peters and Mahan, 2008). The minimum levels of trace minerals required to overcome deficiency syndromes and not necessarily to promote optimum productivity or enhance immunity are listed in the National Research Council (NRC) reviews (1998; 2012). For hyper-prolific modern sows the consequences of an inadequate supply of dietary minerals have been reported on by Mahan and Newton (1995). They showed that, when supplied according to NRC standards, the body mineral

reserves of third parity sows were lower in sows with high than low mean litter weight at 21-days. Furthermore, for either of the latter groups of sows the mineral reserves were lower, by as much as 20%, than those of unbred control animals of similar age. The above explain why NRC (1998) minimum levels are usually exceeded by the industry in order to achieve optimal animal performance (Whittemore et al., 2002). Anil (2011) and Anil et al. (2009; 2010) reported a lower incidence and severity of claw lesions in group housed sows on a diet supplemented with organic zinc, copper and manganese, as a partial substitution of their inorganic form, in comparison with sows on a diet with inorganic trace minerals.

The majority of supplemented minerals in swine feed are used in their inorganic forms as salts (usually oxides or sulphates). However, when conventional inorganic oxides and sulfates (e.g. ZnO or CuSO₄) in feed break down in the stomach, the released ions are free to interact with ligands, which will either allow them to remain soluble in the intestine or bind them to insoluble chelates, like phytate, and form low solubility salts which are unabsorbable (Peters, 2006). As a result, significant proportions of inorganic minerals are fecally excreted (Gerber and Steinfeld, 2008). The valence state of the mineral and its molecular form (inorganic or organic) significantly affect its bioavailability (Miles, 2000). These specific properties of the mineral may be responsible for the complexes they form with other components in the gut, which may either obstruct or facilitate the mucosal absorption, transport and/or metabolism of the mineral in body tissues (Miles, 2000). Minerals joined with an organic ligand such as a protein or a specific amino acid (Carter, 1996), are more soluble and thus can be

absorbed by the intestinal lumen more easily (Anil, 2011) utilizing intestinal uptake processes of amino acids (Power and Horgan, 2000). Wedekind et al. (1992) while aiming to develop bioassay procedures that would allow accurate assessment of zinc bioavailability found that the bioavailability of zinc from zinc methionine (ZnMet) in poultry was greater than that of zinc sulfate. Nockels et al. (1993) showed that except for their bioavailability organic minerals are also better retained by the body. Specifically, he recorded a 62% higher copper retention in calves fed copper lysine (CuLys) than in calves fed copper sulfate (CuSO₄). Similarly, zinc retention was 58.3% higher in calves fed zinc methionine (ZnMet) than in calves fed zinc sulfate (ZnS04), which was a result of better mineral absorption and reduced fecal and urine losses. Regarding the bioavailability of organic trace minerals compared to inorganic ones in swine, there is a controversy. Studies comparing organic Zn sources to Zn sulfate in pigs have indicated higher (Matsui et al., 1996) or equal (Swinkels et al., 1996; Cheng et al., 1998) bioavailability when growth performance and/or tissue concentrations were the response criterion. Similarly, Coffey et al. (1994) and Zhou et al. (1994) reported that Cu-Lysine improved growth performance in pigs whereas liver Cu contents were not affected as compared to Cu sulfate. In another study, copper absorption and retention was greater in weaning pigs when 100 ppm of Cu from a Cu-proteinate was fed, compared to when 250 ppm of Cu sulfate (Veum et al., 2004). In more recent studies conducted in sows, a positive effect of diets supplemented with combinations of organic minerals in sows' reproductive performance (Peters, 2006; Close, 2007; Peters and Mahan, 2008; Anil, 2011) and hoof health was reported (Anil, 2011). This positive effect was attributed to

increased bioavailability (estimates expressed as a percentage of a recognized standard) or alternatively a better absorption and utilization of the complexes of trace minerals. Therefore, using organic mineral sources instead of inorganic ones could help achieve a balance between animal trace mineral needs and their concentration in the diet, without affecting compliance with the limits of their inclusion rate in the feed (Poulsen and Carlson, 2008).

1.4. Foot lesions and reproductive performance

Anil et al. (2009) reported a lower number of liveborn piglets in lame compared to non-lame sows and Pluym et al. (2013b) found that lameness was associated with an increased number of mummified fetuses. However in other studies, lameness was not associated with reduced number of piglets born alive or sows infertility (Heinonen et al., 2006; Willgert, 2011). Associations of specific reproduction parameters with the presence of hoof lesions in sows has also been reported (Bradley et al., 2009; Anil, 2011; Fitzgerald et al., 2012; Pluym et al., 2013b). The litter weight of sows was negatively associated with overgrown toes and dew claws whereas it was positively associated with the total side wall score (Anil, 2011; Fitzgerald et al., 2012). Also, sows with cracks in the outer claw wall and difference in length between lateral and medial toe had increased piglet mortality compared to sows with normal hooves (Fitzgerald et al., 2012). However, piglet mortality of sows with overgrown toes was not higher than that in sows with normal claw length (Enokida et al., 2011; Fitzgerald et al., 2012). The number of piglets weaned per litter was decreased for sows with length differences between the medial and lateral toe and for sows with dew claw and

heel overgrowth and wall lesions (Bradley et al., 2009; Anil, 2011; Fitzgerald et al., 2012). Pluym et al. (2013b) recorded higher odds of stillborn piglets in sows with skin lesions above the claw and white line lesions whereas heel lesions were found to be associated with lower odds of crushed piglets. Moreover, the odds of mummified fetuses decreased when wall cracks were present (Pluym et al., 2013b), in agreement with the findings of Bradley et al. (2009), who recorded a negative association between wall lesions and the number of mummified piglets.

1.5. Background and Objectives

Discussions about the implementation of the EU Directive 2001/88/EC for group housing of pregnant sows increased the concern, among Greek pig producers, for the likely negative effects of group housing on sow longevity and overall herd reproductive performance. Their experience, veterinary consultations and relevant scientific literature supported the anticipated negative results of loose housing on the frequency of hoof lesions and the associated lameness in their sow herds (Anil et al., 2007; Fitzgerald et al., 2012). However, no data were available on the prevalence and severity of claw lesions and lameness in Greek herds, as well as on managerial factors influencing these conditions. Although there is an agreement among studies conducted in Europe and the USA regarding the prevalence of claw lesions in sows (Anil et al., 2007; Enokida et al., 2011; Pluym et al., 2011), it is still unclear if lesions or combinations of lesions on different hoof sites and/or feet affect sow locomotion ability in varying way and degree. Some managerial strategies which were proposed for the reduction of

the incidence of hoof lesions in group housed sows, gave either inconsistent results (i.e. claw trimming) (Kroneman et al., 1992; Vestergaard et al., 2008; Spoolder et al., 2009; DeDecker et al., 2016) or are inappropriate (i.e. deep straw bedding) in the Mediterranean temperate climate of Greece, where the sows should be housed on partially or fully slatted floors. Alterations in the nutritional management of the sows with the use of organic trace minerals gave promising results (Webb et al., 1984; Greer et al., 1991; Anil et al., 2009; 2010; Anil, 2011). When they are supported by strong scientific evidence, nutritional interventions are more easily accepted and applied by the farmers, and may, therefore, have a much faster positive impact on animal welfare and productivity, than investments on flooring or housing, which are usually perceived as longer-term plans. Except for sow longevity, there is still controversy regarding the possible role of hoof lesions on sows' reproductive efficiency (Enokida et al., 2011; Fitzgerald et al., 2012; Pluym et al., 2013b). Clearly, in order for the pig producers to be able to estimate the relevant economic impact and take the correct decisions regarding treatment and/or prevention of sow hoof lesions, their effect on important reproductive parameters should be identified and quantified.

Therefore, the overall goal of the thesis, which comprises studies conducted in three Greek swine herds, was the increase of our understanding about claw lesions as risk factor for sow longevity, productivity and welfare, and the generation of new and update of already reported knowledge regarding the effect of nutritional adjustments on hoof lesions. The goal was served by the following specific objectives:

1) The characterization of sow claw lesions, the estimation of their frequency and severity, and the investigation of their association with parity and

lameness in the studied herds.

2) The investigation of the associations between the severity of claw lesions

and the number of live-born and weaned piglets and the wean-to-first

service interval in the three herds.

3) The investigation of the effect of diet supplementation with chelated zinc

(Zn), copper (Cu) and manganese (Mn) on hoof lesions of loose-housed

sows in the three herds.

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36

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Chapter 2

Hoof lesions and lameness in sows in three Greek swine herds1

M. Lisgara¹, V. Skampardonis¹, S. Kouroupides², L. Leontides¹

¹Department of Epidemiology, Biostatistics and Economics of Animal Production, School of Veterinary Medicine, University of Thessaly, 224 Trikalon st, 43100 Karditsa, Greece.

²VKK Consulting, 65 Iroon Polytechnioy st, 43100 Karditsa, Greece.

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2.1. Summary

Hoof lesions, which are very common among sows, have been associated with

lameness and early culling or euthanasia. In this study, we characterized foot

lesions, estimated their frequency and severity, and investigated their association

with parity and lameness in three Greek farrow-to-finish swine herds. The studied

sows, which had been individually stalled during previous gestations, were

examined for foot lesions upon entry into the lactation facilities. Lesions scored

included heel hyperkeratinization, erosions or cracks, and toe and dew claw

overgrowths. When exiting the farrowing facilities, the sows were observed while

walking along an alley and their degree of lameness was scored. The proportion

of sows with at least one lesion on any foot was very high and similar among

herds, with 121 of 125 (96.8%), 123 of 125 (98.4%), and 377 of 386 (97.7%)

sows affected in herds A, B, and C, respectively. The most frequent lesions were

those located on the heel, and overgrown toes and dew claws. For these sites,

lesion severity increased with sow parity. The concurrent presence of lesions on

more than one foot site, on the same or different feet or both, had a multiplicative

effect on the likelihood of lameness. Under the conditions in the herds

participating in this study, sow foot lesions are extremely common, with older

sows more likely than younger sows to have lesions on the heel and overgrown

toes and dew claws. The degree of lameness may be affected by a causal

interface among foot lesions.

Keywords: swine, hoof lesions, lameness

53

2.2. Introduction

Hoof lesions, an important underlying cause of locomotor disorders in pigs (Dewey et al., 1993), have been associated with lameness and culling or euthanasia (Sanz et al., 2007; Engblom et al., 2008; Jensen and Toft, 2009). Lameness is an animal-based welfare indicator (Welfare Quality® Protocol, 2009). From an economic point of view, lameness reduces the productivity of a pig unit by reducing sow longevity and the number of pigs produced per sow per year due to increased involuntary culling rate of sows, increased expenses as a result of sow replacement costs, increased work load and treatment expenses, and fewer finisher pigs reaching the slaughterhouse (D' Allaire et al., 1987; Anil et al., 2005; Engblom et al., 2007).

In studies conducted in modern herds in the United States and Belgium, almost every sow had at least one foot lesion (Anil et al., 2007; Pluym et al., 2011). However, not all of these sows were lame. Approximately 5% to 20% of lameness cases in sows were attributable to foot lesions (Dewey et al., 1993; Kirk et al., 2005). Location (Anil et al., 2007; 2008) and severity (Gjein and Larssen, 1995a; Pluym et al., 2011) of the lesions might determine whether a sow shows overt lameness or not. Furthermore, hoof injuries may serve as possible ports of entry for infections which may ascend and spread through the body, affecting joints and other tissues, causing stress and pain. Hence, infected hoof lesions can cause severe lameness that reduces the sow's appetite and increases her susceptibility to other diseases, through alteration of the immunological response (Heinonen et al., 2013).

One of the major causes of injuries to the foot at the time of mixing in pens is fighting on concrete or slatted flooring or on combinations of concrete and slatted flooring. Even after the dominance hierarchy is established, grouped sows will continue to fight if they are overstocked, have to compete for access to feed, or are stressed by a perpetual feeling of hunger (Anil et al., 2003; Scott et al., 2009). Previously, fighting was controlled by the use of individual stalls for pregnant sows. However, the European Union (EU) Directive 2001/88/EC, implemented since January 2013 in all 25 member states, requires that sows and gilts be kept in groups during a period starting 4 weeks after service and until 1 week before the expected time of farrowing. Without managerial adjustments, it is reasonable to expect that the importance of foot lesions and associated lameness to longevity and productivity of grouped sows will increase (Anil et al., 2007; Fitzgerald et al., 2012). In this study, conducted in three Greek swine herds during the first 6 months of 2013, sow foot lesions were characterized, their frequency and severity were estimated, and their associations with parity and lameness were investigated. The results depict the baseline prevalence and severity of foot lesions before implementation of the directive for group housing in these herds.

2.3. Materials and methods

2.3.1. Study population

The studied herds were indoor, farrow-to-finish herds with 330 (Herd A), 160 (Herd B), and 800 sows (Herd C), respectively, with Danbred (herds A and B) and Hermitage (Herd C) genotypes. Before finalizing the necessary

reconstruction of the dry-sow units to meet the requirements of the EU Directive 2001/88/EC, all herds kept pregnant sows in individual stalls. Herd C finalized the reconstruction at the end of 2012 and was inspected and granted compliance with the directive by the veterinary authorities in January 2013, whereas herds A and B finalized the reconstructions in the spring and were granted compliance in June 2013.

In the reconstructed units, the animals were loose housed in groups of eight to 12 on combinations of concrete and slatted flooring, as described in the directive. All herds operated on weekly farrowing schedules. Transition to group housing was accomplished gradually, within 4 months after compliance was granted, by grouping the pregnant sows that had been inseminated a month before. Therefore, all sows in the study population had been individually housed during their previous gestations. For participation in the study, the only criterion was the owners' written consent. Neither the health status of the sows' feet nor the frequency of locomotor disorders was considered for herd selection.

2.3.2. Study design

Three farm employees examined the sows' feet for lesions when they entered the lactation facilities. Sow lameness was evaluated upon exit of the animals from the lactation facilities, when managers decided whether a weaned sow would be re-bred or culled, considering reproductive performance, age, and locomotor soundness. Two of the authors (LL and ML) trained the employees to recognize, characterize, and score foot lesions and lameness. An initial training session was held at the clinics of the School of Veterinary Medicine, University of Thessaly (Karditsa, Greece), where the different anatomical sites of the foot were

identified, and representative foot lesions in feet collected at slaughter were characterized and scored. Lameness identification and scoring were demonstrated in a video of sows with normal or abnormal gait and posture. Training was repeated on each farm, and employees were provided with a collection of photographs and the video of the training material. Each sow's data was recorded on especially developed paper data-capture forms. The primary author visited all farms once a month, collected the completed data-capture forms, and cross-checked the data by re-examining a random sample of 20% of the sows with the responsible farm employee. The medial and lateral toes of each foot were individually examined for lesions and scored both when sows were lying down (the ventral surface) and standing up (the dorsal surface) in the farrowing crate before farrowing. Lesions included heel hyperkeratinization. erosions or cracks, and toe and dew claw overgrowth. Specifically, five hoof anatomical sites were examined: the heel (soft keratinized epidermis on the ventral surface of the hoof towards the caudal end); the sole (hard keratinized epidermis cranial to the heel on the ventral surface of the hoof, including the junction between heel and sole); the white line (junction between sole and wall); the wall (hard keratinized epidermis on the dorsal surface of the hoof); and the coronary band. The scoring system applied (Table 2.1 and the Appendix) was based on "Zeugenklauwencheck," a scoring system developed in the Netherlands (Pluym et al., 2011), and the Zinpro Foot First method (Foot First Team, 2010), with some modifications. Epidermal lesions and length of toes and dew claws were scored on a severity scale ranging from 0 to 2, with the exception of the coronary band lesion, where the score was 0 when healthy and

1 when any lesion was observed. On exiting the farrowing facilities, sows were observed from the front and rear while walking down an alley, and their difficulty in bearing weight on one or more feet was scored. Sows exhibiting normal gait were assigned lameness score 0 (non lame); those with alteration or shortening of stride, without serious locomotion impairment or reluctance to move, showing partial inability to bear weight on one or more feet, were assigned score 1; and those with serious locomotion impairment and reluctance to move, showing complete inability to bear weight on one or more feet, were assigned score 2.

Table 2.1. Scoring system applied for evaluation of lesions on seven foot sites of 636 sows in three Greek farrow-to-finish herds*

Foot site	Score 0†	Score 1‡	Score 2§	
Sole	No lesions or very small superficial cracks in the epidermis	Serious lesions in the epidermis not extending into the corium, heel-sole separation, or both	One or more deep cracks extending into the corium, severe heel-sole separation, or both	
Heel	No lesions or very small superficial cracks in the epidermis	Hyperkeratinization and erosions in the epidermis not extending into the corium	Hyperkeratinization, deep cracks extending into the corium, and often necrosis	
White line	No lesions or very small superficial cracks in the epidermis	Wall-sole separation not extending into the corium	Wall-sole separation extending into the corium	
Wall	No lesions or very small superficial cracks in the epidermis	Cracks not extending into the corium, often accompanied by bruising	Cracks extending into the corium, separation of the keratin, or both	
Coronary band	No lesions or very small superficial cracks in the epidermis	Edema with purulent exudate, hemorrhage and necrosis, or both	NA	
Toe	Normal length	Overgrown toes	Overgrown and twisted or cracked toes	
Dew claw	Normal length	Overgrown dew claws, touching the floor when the animal is standing	Overgrown and twisted or crushed dew claws	

- * Based on a Dutch scoring system (Zeugenklauwencheck) and the Foot First Method with some modifications.
- † Corresponding to "score 1 or 2" in the Dutch scoring system or "mild" in the Foot First system.
- ‡ Corresponding to "score 3" in the Dutch system or "moderate" in the Foot First system. For the coronary band, the score applied in this study corresponds to "score 3 or 4" in the Dutch system.
- § Corresponding to "score 4" in the Dutch scoring system or "severe" in the Foot First system.

NA = not applicable; for the coronary band, lesion score was 0 when healthy and 1 when any lesion was observed

2.3.3. Statistical analysis

All statistical analyses were performed using Stata 13.1 (Stata Statistical Software. College Station, Texas). The total score for the four feet for each anatomical site was obtained by adding the respective scores of hooves, toes, and dew claws. Therefore, for each anatomical site except the coronary band, the total score for the four feet could range from 0 to 16; for the coronary band, the total score could range from 0 to 8. The total score for each foot was obtained by adding the scores for each anatomical site considered. Therefore, the total score for each foot could range from 0 to 13.

Subsequently, descriptive statistics of the data were calculated. The Wilcoxon signed-rank test was used to compare the medians of total scores of lesions in each anatomical site between front and rear feet, in each herd. McNemar's χ^2 test for symmetry was used to compare the proportion of sows with at least one lesion in front and rear feet. Pearson's χ^2 test was used to compare the proportion of sows with lesions on each site scored among the three herds, whereas the medians of scores for each site were compared among herds with the Kruskal-Wallis test. Multiple comparisons were interpreted at Bonferroniadjusted P values.

Three ordered logistic regression models were fitted to estimate the association between parity and the total score on all feet, one for each of the three most frequently recorded lesions, which were heel lesions, overgrown toes, and overgrown dew claws. In each model, parity was the dependent variable, while the total lesion score was the explanatory variable. Parity was characterized in one of three categories (parity groups) (PGs) comprising parities

1 or 2 (PG1), 3 to 5 (PG2), and ≥ 6 (PG3). A dummy variable coding for "herd" was forced in all models because it controlled for variation in the outcome due to different herd-parity distribution and unmeasured factors associated with it, as well as different sampling frequency. The assumption of proportionality in the odds did not hold for herd in the models associating parity with heel lesions and dew-claw length, and for toe length in the model associating parity with this lesion. Thus, partial proportional odds models were fitted using the gologit2 command. These models are less restrictive than the parallel-lines models, but more parsimonious and interpretable than those fitted by a non-ordinal method, such as multinomial logistic regression (Williams, 2006).

In herds A and B, the recorded frequency of lame sows was very low, and therefore their data was not considered in the analysis of the association between lameness and severity of foot lesions. Scoring of lesions at the seven foot sites considered resulted in 56 variables for each sow examined. The major problem to be dealt with in analyzing this data set was multicollinearity, ie, predictor variables were closely related to each other (highly correlated) because they referred to the same animal or foot, or even to the same claw. The available techniques to deal with multicollinearity include either exclusion of highly correlated variables after screening for associations among the independent variables, or creation of indices or scores which combine data from multiple factors into a single variable, or creation of a smaller set of independent variables through use of multivariable techniques such as principal components or factor analysis (Dohoo et al., 1997). We opted to conduct factor analysis to consolidate the information contained in all the original variables into a new smaller set of

uncorrelated variables (factors). In factor analysis, the original variables are assumed to be a linear combination of the factors with weights (factor loadings) plus an error term (Dohoo et al., 1997).

Extraction of the factors was accomplished by using the method of principal components (Berghaus et al., 2005). The suitability of individual variables for use in the factor analysis was evaluated by using the Kaiser-Meyer-Olkin measure of sampling adequacy. Determination of the number of factors to keep for interpretation was a compromise between parsimony, interpretability, and the total amount of variation in the original variables that was explained by the factors in the model (Berghaus et al., 2005). Kaiser's criterion (initial eigenvalue ≥ 1), a scree-test plot, and the number of factors that are required to account for a given proportion of the variance observed in the original variables (Stevens, 1996) were considered in the analysis to determine which factors to retain for interpretation. Orthogonal and oblique factor rotations were both evaluated, but ultimately an orthogonal rotation (varimax option) was selected for the final analysis because it resulted in a relatively simple and interpretable structure while maintaining factor independence (Berghaus et al., 2005). Factor loadings > 0.40 were used in the interpretation of rotated factors. Sixteen factors had an eigenvalue ≥ 1, suggesting that they should be kept for interpretation according to Kaiser's criterion, while use of the scree method suggested that 15 or 16 factors should be retained. After consideration of the amount of variance explained, we retained 16 factors, cumulatively accounting for almost 70% of the variance in the original variables. Then, for these 16 factors, the regression method was used to produce standardized factor scores (Berghaus et al., 2005).

Subsequently, the produced standardized factor scores were evaluated as predictors of lameness score in an ordinal logistic regression model. Adjustment for the likely parity effect was accomplished by forcing parity into the model (Willgert et al., 2014). Because the assumption of proportionality did not hold for all predictors examined, we fitted partial proportional odds models (Williams, 2006). To identify partial proportional odds models that fitted our data best, we used the autofit option, which is a built-in option of gologit2. When this option is specified, gologit2 goes through an iterative process. Initially it fits a totally unconstrained model and then performs a series of Wald tests on each variable to determine whether its coefficients differ across equations, eg, whether the variable meets the parallel-lines assumption. If the test is significant for one or more variables, the variable with the least significant value is constrained to have equal effects across equations. The model is then refitted with constraints, and the process is repeated until there are no more variables that meet the parallellines assumption. Finally, a global Wald test is done, which compares the final model with constraints to the original unconstrained model and, if the Wald test is statistically insignificant, the final model does not violate the parallel-lines assumption (Williams, 2006).

For factor score selection for the final regression model, we initially fitted bivariable models, including each factor score and parity. Factor scores significant at P < 0.25 were candidates for the final model (Hosmer et al., 1989). The initial full model fitted included parity and all standardized factor scores previously identified as significant. It was then reduced by backwards elimination of factor scores with $P \ge 0.05$ (Mickey and Greenland, 1989). When only those

with *P* < 0.05 remained, factor scores previously eliminated were offered one at a time to the model. This ensured that factor scores excluded earlier, during backward elimination, but adding significantly to the final model, were not missed. Lastly, all possible two-way interactions between factor scores in the model were created and tested for significance one by one. The fit of the final model to the data was assessed by comparing the observed to model-predicted probabilities of occurrence of each lameness score (Rabe-Hesketh and Skrondal, 2008).

2.4. Results

2.4.1. Foot lesions

A total of 636 sows were scored, of which 125 were in Herd A, 125 in Herd B, and 386 in Herd C (Table 2.2). The proportion of sows with at least one lesion on any foot was very high and similar among herds with 121 of 125 (96.8%), 123 of 125 (98.4%), and 377 of 386 (97.7%) affected sows in herds A, B, and C, respectively. In Herd C, the proportion of sows with at least one lesion on the front feet (338 of 386; 87.6%) was lower (P < 0.001) than the proportion of sows with at least one lesion on the rear feet (378 of 386; 97.9%). However, these proportions did not differ in Herd A or Herd B.

The most frequent and severe foot lesions observed in each herd separately are shown in Table 2.2. There was among-herd variation in the frequency and severity of these lesions. Heel lesions were less frequent (*P* < 0.001) in Herd A than in Herd B or Herd C. Frequency of heel lesions did not

differ (P = 0.10) between Herd B and Herd C. The total score of heel lesions differed (P < 0.001) among the three herds, being lowest in Herd A and highest in Herd C. Both the frequency and severity of overgrown toes differed among the herds ($P \le 0.001$ in each comparison), being more frequent and severe in Herd A and least frequent and severe in Herd C. Similarly, the frequency and severity of overgrown dew claws differed among the three herds (P < 0.001 in each comparison), being more frequent in Herd A and more severe in Herd C, and least frequent and severe in Herd B. In general, within herds, the median scores of the heel lesions, toe, and dew-claw length were higher (P < 0.02 in each comparison) for the rear than for the front feet, with the exception of the toe length in Herd A sows, which did not differ between front and rear feet (P = 0.29).

Table 2.2. Frequency of sows with at least one foot lesion and median (range) of the total score* for all feet by site and herd in a study conducted in three Greek farrow-to-finish herds

Lesion frequency by site on the foot							
Herd	Sole	Heel	White line	Wall	Coronary	Toe length	Dew-claw
	(%)	(%)	(%)	(%)	band (%)	(%)	length (%)
A	55	65	58	67	21	115	114
n = 125	(44.00)	(52.00)	(46.40)	(53.60)	(16.80)	(92.00)	(91.20)
В	70	112	24	84	12	96	83
n = 125	(56.00)	(89.60)	(19.20)	(67.20)	(9.60)	(76.80)	(66.40)
С	207	362	148	212	63	162	322
n = 386	(53.63)	(93.78)	(38.34)	(54.92)	(16.32)	(41.97)	(83.42)
			Median of tot	al score (range)		
A	0 (0-13)	1 (0-12)	0 (0-8)	1 (0-11)	0 (0-8)	4 (0-14)	3 (0-15)
В	1 (0-7)	2 (0-10)	0 (0-6)	1 (0-4)	0 (0-3)	2 (0-7)	2 (0-9)

C 1 (0-12) 7 (0-10) 0 (0-11) 1 (0-10) 0 (0-5) 0 (0-10) 5 (0-16)

^{*} Scores defined in Table 2.1. The total score for the four feet for each anatomical site was obtained by adding the respective scores of hooves, toes, and dew claws.

2.4.2. Association of lesions with parity

For heel lesions and for overgrown toes and dew claws, which were the most common lesions, there were associations of parity with the total score (Table 2.3). These associations were adjusted for the herd effect, which was included in the models as a confounder. For each unit increase in the total score of heel lesions and dew-claw length, a sow was 1.10 times (P < 0.001) and 1.20 times (P < 0.001) more likely, respectively, to belong to PG2 or PG3 than to PG1. Additionally, for each unit increase in the total score of toe length, a sow was 1.15 and 1.26 times more likely (P < 0.001) to belong to PG2 or PG3 than to PG1 and to PG3 than to PG1, respectively.

Table 2.3. Odds ratios (OR) and 95% confidence intervals (CI) for herd-adjusted associations between sow parity group (PG)* and total lesion score on heel, overgrown dew claws, and overgrown toes.

Foot site	PG ≥ 2 versus PG1 and PG3	PG ≥ 2 versus PG1	PG3 versus PG ≤ 2	
	versus PG ≤ 2			
-	OR (95% CI)	OR (95% CI)	OR (95% CI)	
Heel†	1.10 (1.06-1.14)	NA	NA	
Overgrown dew	1.20 (1.19-1.26)	NA	NA	
claws†				
Overgrown toes‡	NA	1.15 (1.06-1.20)	1.26 (1.07-1.40)	

^{*} PG1, parity 1 or 2; PG2, parities 3-5; and PG3, parities ≥ 6.

[†] The assumption of proportionality in the odds is valid.

[‡] The odds ratios are not constant across PGs because the assumption of proportionality in the odds is not valid. NA = not applicable.

2.4.3. Association of lesions with lameness

In Herd C, the proportion of sows with locomotor disorders was 81 of 386 (21.0%). Specifically, 53 of 386 (13.7%) and 28 of 386 (7.3%) sows had lameness scores 1 and 2, respectively. In herds A and B, three of 125 and one of 125 sows, respectively, had lameness score 1, whereas none had lameness score 2.

All variables examined were suitable for inclusion in the factor analysis, since their Kaiser-Meyer-Olkin values were > 0.5, suggesting an acceptable fit with the structure of the other variables. Most variables loaded high on only a single factor, the exception being Factor 1. For this factor, three different groups of variables loaded: variables describing scores of white-line and sole lesions and of toe length of the rear foot. During final model building, five factor scores were found significant after backwards elimination, and another during forward selection. None of the examined interactions were significant. Thus the final model included factor scores 1, 2, 5, 7, 11, and 13 as independent variables (Box 2.1).

According to the final model, lameness was associated with lesions on five foot sites. For all but one site, lameness severity increased with increasing lesion score, the exception being the wall of the front hoof, factor score 11 (Table 2.4). The likelihood of lameness score being \geq 1 compared to 0 was almost three times higher (P < 0.001) per one unit increase in factor score 1, whereas it was almost two times higher (P < 0.001) for lameness score 2 compared to \leq 1. For one unit increase in factor score 2, the odds were 1.90 times higher (P = 0.004) that lameness score would be 2 rather than \leq 1. It was 1.70 times more likely (P = 0.004)

< 0.001) that a sow would have a higher lameness score for a unit increase in factor score 5. It was 1.40 times more likely (P = 0.005) that a sow would have a higher lameness score for a unit increase in factor score 7. It was 1.50 times more likely (P = 0.001) that a sow would have a higher lameness score for a unit increase in factor score 13. It was 0.60 times less likely (P = 0.006) for a sow to be lame for a unit increase in factor score 11.

Box 2.1: Factor scores* included in the final model for lameness, representing the lesion scores† for the foot sites examined in 386 sows

Factor score 1, for toe length and white-line and sole lesions, rear hooves

Factor score 2, for dew-claw length, front hooves

Factor score 5, for dew-claw length, rear hooves

Factor score 7, for toe length, front hooves

Factor score 11, for wall lesions, front hooves

Factor score 13, for white-line lesions, front hooves

^{*} Scoring of lesions at the seven foot sites considered (Table 2.1) resulted in 56 variables for each sow examined. From these multicollinear variables, a smaller set of independent variables (factors) were extracted using factor analysis. The regression method was used to produce factor scores for these factors.

[†] Scores defined in Table 2.1.

Table 2.4. Odds ratios (OR) and 95% confidence intervals (CI) for parity-adjusted associations between factor scores and lameness for 386 sows in one Greek farrow-to-finish herd (Herd C)*

Factor scores	Lameness score ≥ 1 versus 0	Lameness score ≥ 1 versus	Lameness score 2 versus score ≤ 1	
	and score 2 versus score ≤ 1	score 0		
	OR (95% CI)	OR (95% CI)	OR (95% CI)	
1†	NA	2.90 (2.10-4.00)	1.80 (1.30-2.40)	
2†	NA	1.20 (0.90-1.60)	1.90 (1.02-3.00)	
5‡	1.70 (1.30-2.30)	NA	NA	
7‡	1.40 (1.10-1.70)	NA	NA	
11‡	0.60 (0.40-0.80)	NA	NA	
13‡	1.50 (1.20-1.90)	NA	NA	

^{*} Factor scores defined in Box 2.1. Lameness scored from 0 (non-lame) to 2 (complete inability to bear weight on one or more feet).

NA = not applicable.

[†] The odds ratios are not constant across lameness scores because the assumption of proportionality in the odds is not valid.

[‡] The assumption of proportionality in the odds is valid.

2.5. Discussion

This study is part of a greater project aiming to characterize foot health and improve sow longevity in Greek swine herds with managerial and nutritional interventions. In the first part of the project, presented here, we estimated the frequency and severity of foot lesions and associated lameness in three herds with general management and housing typical of that in most Greek herds. We initially developed and documented a scoring system for lesions and lameness which was similar to those used in previous reports, with some modifications. Almost every sow examined in the three herds had at least one lesion, and the most frequent and severe were the heel lesions and the overgrown toes and dew claws. Likewise, other studies also recorded an extremely high frequency of foot lesions in sows (Anil et al., 2007; Pluym et al., 2011). Heel lesions and hoof wall cracks were the most common (Gjein and Larssen, 1995b; Anil et al., 2007; Enokida et al., 2011; Grégorie et al., 2013) whereas the most severe lesions were detected on the heel and the dew claws (Pluym et al., 2011). We found a positive association between parity and severity of lesions. Older sows were more likely to have severe heel lesions and overgrown toes and dew claws. Hoof abnormalities occurred more frequently and were more severe in older sows (Dewey et al., 1993; Pluym et al., 2011; Fitzgerald et al., 2012) although a reverse effect has also been reported (Anil et al., 2007), probably due to the differential culling rate of affected sows. Since the heel bulb, mainly of the lateral digits, carries most of the sow's weight (Webb et al., 1984), and high-parity sows, on average, weigh more than younger sows, the heel area is stressed more in older than younger sows. Furthermore, the mean rate of hoof horn growth in

sows was recently estimated at approximately 6.3 mm and the mean wear rate at approximately 5.1 mm per month (Amstel and Doherty, 2010). Therefore, toe overgrowth may occur simply as a function of age, especially when sows are not provided with enough space for exercise. Formation of hoof horn is a complex and structured process of cellular changes that transform living, highly functional epidermal cells into mechanically very stable horn cells. This process of horn formation is sensitive to nutritional influences, hormones, and environmental factors (Muelling, 2009; Van Riet et al., 2013).

In general, lameness is considered a multifactorial phenomenon with several physiological causes (infectious and non-infectious) affecting various tissues and anatomical structures (Nalon et al., 2013). There is evidence that some types of foot lesions cause lameness and poor reproductive performance (Pluym et al., 2011; Fitzgerald et al., 2012). The link between foot lesions and lameness is believed to be pain mediated (Bonde et al., 2004). Typically, the location (Anil et al., 2007) and severity of lesions (Gjein and Larssen, 1995a) are important factors. However, several relevant studies have either failed to demonstrate a significant association (Kroneman et al., 1993) or identified few specific foot lesions (ie, white-line lesions, overgrown toes) associated with lameness (Hulten et al., 1995; Jørgensen, 2000; Anil et al., 2007). In our attempt to associate foot lesions with lameness, we employed factor analysis, which handled the limitations and complications involved in the simultaneous evaluation of a large number of variables, many of which were correlated. We were able to identify a causal interface between various foot lesions and lameness scores. Some lesions affected lameness scores when they were combined (factor score

1), whereas others had a discerned effect according to their location. Lesions located on five sites of the foot, namely the white line, sole, wall, and overgrown toes and dew claws, were associated with lameness. Furthermore, the concurrent presence of lesions on more than one foot site, on the same or a different foot had a multiplicative effect on the likelihood of lameness. It is understandable that severe white-line and sole lesions can affect some gait parameters in sows (Gregorie et al., 2013). Since the white line is the junction of wall and sole horn, injuries on that site may easily facilitate the invasion of bacteria into the corium, causing pain and inflammation. This can lead to locomotor disorders in sows (Anil et al., 2007) and in cows – white-line disease (Kempson and Logue, 1993). According to the experience obtained in this study, lesions on the white line of a hoof were frequently accompanied by lesions on the sole, since these two sites are adjoined. The prominent clinical sign of locomotion disorder associated with long toes was a gait abnormality that has been described as "goose-stepping of rear legs" (Jørgensen, 2000). Severe overgrowth of toes and dew claws was associated with lameness (Hulten et al., 1995; Pluym et al., 2011) and was reported to be the most common foot lesion responsible for culling (Dewey et al., 1993). When sows are kept on fully or partially slatted floors, overgrown toes and dew claws may be caught between slats. When the animal attempts to move they may be cracked, and dew claws especially may be completely ripped off. Furthermore, overgrown dew claws may be concave and extend beneath the heel bulb, which is thereby traumatized. Thus, bacteria can enter the corium, causing infection and pain. These observations may explain why sows with long dew claws were more likely to be

lame. Therefore, regular trimming of dew claws, which grow along with the toes but do not normally touch the ground to wear, may be a valuable measure to mitigate the risk of lameness (Zinpro Foot First).

The results of our study are limited to the extent that recording and scoring of lesions and lameness were conducted by farm personnel. Although there were training sessions for lesion characterization and lameness diagnosis by the personnel, and the validity of a subsample of the recordings was verified by one of us (ML), there were differences among herds. These differences were due not only to the unavoidable imperfect validity and repeatability of personnel scorings, but also to the existing variations in management, productivity, and genetic lines of sows. In two of the three herds (herds A and B), primarily managerial decisions for quick culling of sows with locomotor problems, and secondarily limited ability to detect lame sows, resulted in very low frequencies of lame sows. Using the data from the third herd, we identified significant associations between several foot lesions and lameness score. Our analytical approach was able to identify groups of closely related foot lesions among a larger set of 56 variables describing lesions on the feet of each sow, without losing any important information, and minimizing the possibility of finding associations "due to chance alone" (Dohoo et al., 1997). We showed that the degree of lameness was affected by a causal interface among various foot lesions. Although generalization of these results is risky, since the data originated from one herd, when combined with the results of other studies (Pluym et al., 2011) they point out the need for general improvement in foot health. Though housing conditions and management on the farm are crucial as immediate causes for development

of foot lesions (Kroneman et al., 1993), trace-mineral nutrition should also be considered a predisposing factor, because it is vital in developing foot structure and integrity (Tomlinson et al., 2004). Proper nutrition with supplementation of proteinated trace mineral may improve the quality of the hoof horn tissue and reduce its susceptibility to chemical, physical, or microbial damage from the environment (Tomlinson et al., 2004). It should, therefore, very likely be part of managerial changes required for transition from individual to loose housing of pregnant sows.

2.6. Conclusions

Under the conditions of this study in three Greek herds, sow foot lesions are extremely common. Also, older sows are more likely than younger sows to have heel lesions and overgrown toes and dew claws and the degree of lameness in sows may be affected by a causal interface among foot lesions.

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83

Chapter 3

Associations between claw lesions and reproductive performance of sows in three Greek herds²

M. Lisgara¹, V. Skampardonis¹, E. Angelidou¹, S. Kouroupides², L. Leontides¹

¹Department of Epidemiology, Biostatistics and Economics of Animal Production, School of Veterinary Medicine, University of Thessaly, 224 Trikalon st, 43100 Karditsa, Greece.

²VKK Consulting, 65 Iroon Polytechnioy st, 43100 Karditsa, Greece.

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3.1. Summary

Claw lesions, which are nowadays very common in sows, are associated with a high risk of early culling and compromised welfare. In this study, we investigated the associations between the severity of claw lesions and three of the most important reproductive indicators, the number of live-born and weaned piglets and the wean-to-first service interval in three Greek farrow-tofinish herds. All studied sows were individually housed during their previous gestations. Sows were examined for lesions, which were scored on a severity scale, on several anatomical sites of the claws, before farrowing. Data on the examined reproductive indicators were retrieved from productivity databases of the herds. Because scoring of lesions on several claw sites resulted in many correlated variables for each sow examined, we employed factor analysis to create a smaller set of uncorrelated variables (factors) which contained all the information in the original variables and produced the corresponding factor scores. The number of live-born and weaned piglets was associated with the produced factor scores in two multivariable linear regression models, whereas the possible association between the wean-tofirst service interval and the factor scores was modelled with the use of zeroinflated negative binomial regression. The number of live-born piglets was negatively associated with factor scores representing lesions on heel (P≤ 0.001) and sole of front feet (P = 0.019). The number of weaned piglets was also negatively associated with factor scores representing lesions on heel (P = 0.003) of any foot, on sole of front feet (P = 0.001) and on white line, sole and wall of rear feet (P = 0.008), while the wean-to-first service interval was associated with factor scores representing lesions on heel of any foot (P =

0.02), on sole of front feet (P = 0.02) and of dew claw length of front feet (P = 0.009). Our results indicate that combinations of lesions on the dorsal and ventral part of the claws, negatively affected the reproduction parameters considered, emphasizing the importance of general improvement of feet

Keywords: sows; claw lesions; reproductive performance

3.2. Introduction

health.

Claw lesions, which are very common among sows (Pluym et al., 2011), have been associated with lameness (Anil et al., 2007; Lisgara et al., 2015).

Lameness is an animal-based welfare indicator (Welfare Quality® Protocol, 2009) which reduces the productivity of a pig herd by reducing sow longevity and the number of pigs produced per sow per year due to increased involuntary culling rates of sows (Anil et al., 2005; Engblom et al., 2008).

Some claw lesions were also associated with decreased litter weight, increased pre-weaning piglet mortality and higher odds of stillborn and crushed piglets (Fitzgerald et al., 2012; Pluym et al., 2013).

A commonly discussed parameter for determining breeding herd reproductive performance and a herd's overall efficiency is pigs weaned/female/year. This parameter is principally influenced by the non-productive days/female/year, which is in-turn influenced by the wean-to-first service interval, the total number of piglets born and weaned and the lactation length (Polson et al., 1993; Almond et al., 2006). Evidently, if claw lesions negatively affect not only sow longevity but also the important reproductive parameters which determine the breeding capacity of the herd and the

profitability of the farm, every effort should be made to combat this problem.

Thus, in this paper, we investigated the associations between the severity of claw lesions and the number of live-born and weaned piglets and the wean-to-first service interval in three Greek farrow-to-finish herds.

3.3. Materials and methods

3.3.1. Study population

The studied herds were, indoor, farrow-to-finish herds with 330 (A), 160 (B) and 800 (C) sows, respectively, with Danbred (A, B) and Hermitage (C) genetics. The only criterion for participation in the study was the written consent of the owners. Neither the health status of sow feet nor reproductive performance was considered for herd selection. The study was conducted during the first six months of 2013, before full compliance with the EU Directive (2001/88/EC), which requires that sows and gilts shall be kept in groups during a period starting from four weeks after service to one week before the expected time of farrowing. Therefore, all sows in the study population were individually housed during all their previous gestations.

3.3.2. Study design

Upon entry of sows into the lactation facilities, their hooves were examined for lesions and scored by three farm employees. The training of the employees to recognize, characterize and score feet lesions was done by two of the authors of this study (ML and LL). Training involved an initial session at the Clinics of the School of Veterinary Medicine of the University of Thessaly (Karditsa, Greece), where the different anatomical sites of the claws were identified and representative claw lesions collected in the slaughterhouse were

characterized and scored. Training was repeated on each farm. For referencing, the employees were provided with a collection of pictures and a video of the training material. Each sow's feet data were recorded on specially developed sheets (paper data-capture forms) along with her parity, date of farrowing, number of live-born and weaned piglets and the wean-to-first service interval. Once a month the primary author visited all farms, collected the sheets and cross-checked the data of a random sample of 20 percent of the sheets by rescoring sow claws together with the responsible farm employee. The medial and lateral toes of each foot were individually examined for lesions and scored when sows were lying down (the ventral surface) or standing up (the dorsal surface) in the farrowing crate before farrowing. Five claw anatomical sites were examined, the heel (soft keratinized epidermis on the ventral surface of the claw towards the posterior end, HL), the sole (hard keratinized epidermis anterior to the heel on the ventral surface of the claw including the junction between heel and sole, SL), the white line (junction between sole and wall, WL), the wall (hard keratinized epidermis on the dorsal surface of the claw, WA) and the coronary band (CB). The scoring system applied was based on "Zeugenklauwencheck", a scoring system developed in The Netherlands (Pluym et al., 2011) and the Zinpro Foot First method (Foot First Team, 2010), with some modifications. Scoring of lesions of the epidermis involved a severity scale ranging from 0 to 2 where score 0 was given to claws with no lesions or very small superficial cracks of the epidermis ("score 1 or 2" in the Dutch scoring system or "mild" in the Foot First system), score 1 was assigned to serious lesions in the epidermis not extending into the corium ("score 3" in the Dutch scoring system or "moderate" in the Foot First system) and score 2 was assigned to severe lesions with serious and

deep cracks extending into the corium or subcutis ("score 4" in the Dutch scoring system or "severe" in the Foot First system). For toe length (TL) and dew claw length (DCL), score 0 was assigned to toes and dew claws with normal length ("score 1 or 2" in the Dutch scoring system or "mild" in the Foot First system), score 1 to overgrown toes and dew claws touching the floor when the animal was standing ("score 3" in the Dutch scoring system or "moderate" in the Foot First system), and score 2 to overgrown and twisted or cracked toes and overgrown and twisted or crushed dew claws ("score 4" in the Dutch scoring system or "severe" in the Foot First system). For the coronary band (CB), lesion score was 0 when healthy ("score 1 or 2" in the Dutch scoring system) and 1 when any lesion was observed ("score 3 or 4" in the Dutch scoring system).

3.3.3. Statistical analysis

All statistical analyses were performed using Stata 13.1 (Stata Statistical Software. College Station, TX). The total score for the four feet for each anatomical site was obtained by adding the respective scores of all claws and dew claws. Therefore, for all anatomical sites except the coronary band, the total score for the four feet could range from 0 to 16; for the coronary band, the total score varied between 0 and 8. The total score for each foot was obtained by adding the scores of each anatomical site considered. Therefore, the total score of each foot could range from 0 to 13. Initially the data were summarized by calculation of frequencies, medians, ranges and means ± SD. Analysis of variance (ANOVA) was used to compare the mean number of liveborn and weaned piglets among the three herds, whereas the medians of the wean-to-first service interval were compared among the herds using the

Kruskal- Wallis test (multiple comparisons were interpreted at Bonferroni adjusted *P*-values).

Scoring of lesions at the considered foot sites resulted in fifty six variables for each sow examined. The major problem to be dealt with in analyzing this data set was multicollinearity, i.e., variables were closely related to each other (highly correlated) because they referred to the same animal and/ or same foot and/or same claw. The first approach when dealing with multicollinearity involves reducing the number of independent variables prior to investigating associations with the outcome. This could be accomplished by screening for multicollinearity and selecting predictor variables, by creating scores or indexes combining data from several variables or by using multivariable methods (principal component analysis or factor analysis) to summarize the information contained in the original predictors into a smaller set of variables (Dohoo et al., 1997). The first alternative is limited by the arbitrary nature of selecting the level of correlation coefficient and which of the correlated variables to remove, while the second precludes the evaluation of the effects of the individual variables which make up the score or index in analyses of risk factors. Between the use of principal component and factor analysis, we opted to use the latter because principal components are merely mathematical constructs with no intrinsic meaning, whereas factor analysis is based on the assumption that a set of factors which do have an inherent meaning of their own can be computed as weighted sums of the original variables (Dohoo et al., 1997). Thus, through the use of factor analysis we created a smaller set of uncorrelated predictor variables (factors) which contained all the information in the original variables. The original variables were assumed to be linear combinations of the factors with

weights (factor loadings) plus an error term. Extraction of the factors was accomplished by using the method of principal components (Berghaus et al., 2005). The suitability of individual variables for use in the factor analysis was evaluated by using the Kaiser-Meyer-Olkin (KMO) measure of sampling adequacy. Determination of the number of factors to keep for interpretation was a compromise between parsimony, interpretability, and the total amount of variation in the original variables that was explained by the factors in the model (Berghaus et al., 2005). Kaiser's criterion (initial eigenvalue ≥ 1), a Scree-test plot and the number of factors that are required to account for a given proportion of the variance observed in the original variables (Stevens, 1996), were considered in the analysis to determine which factors to retain for interpretation. Orthogonal and oblique factor rotations were both evaluated, but ultimately an orthogonal rotation was selected for the final analysis because it resulted in a relatively simple and interpretable structure while maintaining factor independence (Berghaus et al., 2005). |Factor loadings| > 0.40 were used in the interpretation of rotated factors. Sixteen factors had an eigenvalue ≥ 1, suggesting that they should be kept for interpretation according to Kaiser's criterion, while use of the Scree method suggested that 13 or 14 factors should be retained. After consideration of the amount of variance explained, we retained 13 factors cumulatively accounting for almost 63 percent of the variance in the original variables. Then, for these 13 factors we produced standardized factor scores with the regression method (Berghaus et al., 2005).

The number of live-born and weaned piglets was associated with the produced factor scores in two multivariable linear regression models.

However, the distribution of the observed days of wean-to-first service was

over-dispersed (Lord et al., 2007). This was expected because most sows were inseminated within five days after weaning. Thus, we used a zero inflated negative binomial (ZINB) model to investigate the possible associations between the wean-to-first service interval and the factor scores (Carrel et al., 2010). The latter model fitted the data better than the Poisson or negative binomial regression models as dictated by two tests, namely the Vuong and the Countfit tests which are available in Stata 13.1. The ZINB model generates two separate models and then combines them; first, a logit model was generated for sows which were serviced within five days after weaning and then, a negative binomial model was generated for sows which were serviced more than five days after weaning (Long and Freese, 2006).

In all models, adjustment for the likely parity-effect was accomplished by forcing a dummy variable for "parity" (categorized in one of three categories comprising parities 1 or 2, 3 to 5, and \geq 6) into the models. A dummy variable coding for "herd" was also forced into all models as it controlled for variation in the outcomes because of the different within-herd sampling frequency and other unmeasured herd level factors.

The model building procedure was similar for the linear regression and ZINB models. Initially, for each outcome, each factor score was screened separately, together with "parity" and "herd", in tri-variable models. A significance level of P < 0.25 was used as a screening criterion for possible inclusion in the initial full models (Hosmer and Lemeshow, 1989). Then, the initial full models including all factor scores previously found significant at P < 0.25 along with "parity" and "herd" were fitted. Examination of the residuals of the multiple regression models for the number of live-born and weaned piglets did not reveal any notable deviation from normality and homoscedasticity. The

full models were reduced by backwards elimination of factor scores with $P \ge 0.05$ (Mickey and Greenland, 1989). When only significant factor scores remained in the models, a stepwise forward selection process was performed, offering to the models previously deleted factor scores one at a time. This ensured that any factor scores excluded earlier during backward elimination, but adding significantly to the final models, were included. Lastly, all possible two-way interactions between the remaining factor scores were created and tested for significance one-by-one.

3.4. Results

A total of 804 sows were scored, of which 121 were in herd A, 126 in herd B, and 557 in herd C. In herd A, the most frequent and severe lesions were the TL 111/121 (91.7%), median 4 (range 0–14) and the DCL 110/121 (90.9%), median 3 (range 0–15); in herd B they were on the HL 113/126 (89.7%), median 2 (range 0–10) and the TL 97/126 (77%), median 2 (range 0–7); and in herd C they were on the HL 502/557 (90.1%), median 6 (range 0–16) and the DCL 433/557 (77.7%), median 4 (range 0–16).

In herd A, the mean number of live-born and weaned piglets were 14.14 (\pm 3.00) and 12.60 (\pm 1.27); in herd B, they were 14.41 (\pm 2.80) and 11.11 (\pm 1.90); and in herd C they were 10.97 (\pm 2.70) and 10.16 (\pm 1.40), respectively. Means of live-born piglets were lower (P < 0.01) in herd C but did not differ between the other herds, whereas means of weaned piglets differed (P < 0.01) among herds. Some of the sows were culled at weaning; therefore, the wean-to-first service interval was available for 95, 109 and 500 sows in herds A, B and C, respectively. It had a median of six days (range 5–47 days) in herd A, five days (range 3–35 days) in herd B and five days

(range 4–98 days) in herd C; the distributions of days from wean-to-first service differed (P < 0.01) among herds.

All variables examined were suitable for inclusion in the factor analysis, since their KMO values were > 0.5, suggesting an acceptable fit with the structure of the other variables. Most variables loaded high on only a single factor, the exception being factor 10. For this factor, three different groups of variables loaded: variables describing the severity of lesions on the white line, sole and wall of the rear feet. None of the examined interactions was significant. The final model for live-born piglets included factor scores 1, 2 and 4, the one for weaned piglets included factor scores 1, 2, 4 and 10 and the model for wean-to-first service interval included factor scores 1, 2, 4 and 7.

Factor score 1, representing the severity of HL lesions on front and rear feet.

Factor score 2, representing TL on front and rear feet.

Factor score 4, representing the severity of SL lesions on front feet.

Factor score 7, representing DCL on front feet.

Factor score 10, representing the severity of WL, SL and WA lesions on rear feet.

The number of live-born piglets was negatively associated with factor scores 1 ($P \le 0.001$) and 4 (P = 0.019). Specifically, for one unit increase of the former or the latter, a sow farrowed on average 0.36 and 0.24 less piglets, respectively. In contrast, the number of live-born piglets was positively ($P \le 0.001$) associated with factor score 2; for one unit increase of factor score 2 the number of live-born piglets increased by an average of 0.38 piglets (Table 3). The number of weaned piglets was negatively associated with factor

scores 1 (P = 0.003), 4 (P = 0.001) and 10 (P = 0.008), and positively associated with factor score 2 (P = 0.004). On average, a sow weaned 0.17 piglets less for one unit increase of each of the first two factor scores and 0.15 piglets less for one unit increase of factor score 10; for a unit increase of factor score 2 the number of piglets weaned increased on average by 0.16 piglets (Table 3).

In the negative binomial part of the ZINB model, the wean-to-first service interval was associated with factor scores 1 (P = 0.02), 2 ($P \le 0.001$), 4 (P = 0.02) and 7 (P = 0.009). Therefore, for sows serviced more than five days from weaning, one unit increase of either of factor scores 1, 4 and 7 increased the wean-to-first service interval by a factor of 1.30. In contrast, for one unit increase of factor score 2 the wean-to-first service interval decreased by a factor of 0.60. In the logit part of the ZINB model, the wean-to-first service interval was not associated with any factor score.

Table 3. Linear regression models for associations between the number of live-born and weaned piglets and claw lesions of 804 sows, adjusted for parity and herd effects, in three Greek herds, together with the negative binomial part of a zero-inflated negative binomial regression model for the association between the longer than five days after weaning-to-first service interval and claw lesions of 704 sows from the same herds, adjusted for parity and herd effects

Factor scores	es Number of live-born piglets		Number of weaned piglets		Wean-to-first service interval	
	Coefficient (CI)	<i>P</i> -value	Coefficient (CI)	<i>P</i> -value	Coefficient (CI)	<i>P</i> -value
1 ^a	-0.36 (-0.58, -0.16)	≤ 0.001	-0.17 (-0.28, -0.06)	0.003	0.27 (0.04, 0.50)	0.02
2 ^b	0.38 (0.19, 0.60)	≤ 0.001	0.16 (0.05, 0.28)	0.004	-0.47 (-0.69, -0.25)	≤ 0.001
4 ^c	-0.24 (-0.41, -0.01)	0.019	-0.17 (-0.28, -0.07)	0.001	0.26 (0.04, 0.47)	0.02
7 ^d	_	_	_	_	0.28 (0.07, 0.48)	0.009
10 ^e	-	-	-0.15 (-0.26, -0.04)	0.008	-	-

^afactor score representing the severity of heel lesions on front and rear feet

^bfactor score representing the overgrowth of toes on front and rear feet

^cfactor score representing the severity of sole lesions on front feet

^dfactor score representing the overgrowth of dew claws on front feet

efactor score representing the severity of white line, sole and wall lesions on rear feet

3.5. Discussion

In this study we scored lesions on five claw sites, namely, the heel, the sole, the white line, the wall and the coronary band, the toe and the dew claw length. Then, we investigated the associations between claw lesion scores and three main reproductive parameters, the number of live-born and weaned piglets and the duration of the wean-to-first service interval, in three Greek swine herds. We identified lesions located on six foot sites, namely the heel, sole, white line, wall and the overgrown toes and dew claws which were associated with reproductive performance parameters. With the sole exception of the overgrown toes, any increase in the severity score of lesions on the other foot sites had a negative effect on the reproductive parameters considered (Table 3).

Although the impact of claw lesions on sow longevity, lameness (Anil et al., 2007; Anil et al., 2009) and increased risk of culling (Engblom et al., 2008) is established, there are only sparse reports on their associations with reproduction efficiency parameters. Previous studies reported the association of white line lesions and wall cracks with decreased litter weight, increased pre-weaning piglet mortality and higher odds of stillborn piglets (Fitzgerald et al., 2012; Pluym et al., 2013). Our results indicate that combinations of lesions on the dorsal and ventral part of the claws, negatively affected the reproduction parameters considered. These lesions are extremely common (Anil et al., 2007; Pluym et al., 2011) with 56% of the sows, in our study population, having at least two lesions on the ventral part and 58% at least one lesion on the dorsal part of the claw of any foot.

According to our experience, lesions on the white line were frequently accompanied by lesions on the sole of the claw, since these two sites are

adjoined. The heel bulb, mainly of the lateral claws, is a soft tissue prone to injuries because it carries most of the sow's weight (Kroneman et al., 1993; Pluym et al., 2011). All lesions on the ventral or dorsal part of the claw may facilitate the invasion of bacteria into the corium resulting in inflammation and pain, causing, due to cytokine release, anorexia and lethargy (Kempson and Logue, 1993; Johnson, 1997). In our study, the great majority of the animals affected with severe lesions (lesion score 2) had, when culled and pathologically examined, evidence of laminitis (data not shown). Reduced feed consumption as a sequel of claw lesions could be either or both the result of an existing inflammatory process or an impaired locomotor ability. In addition, the postural behavior of the sow may be influenced, with affected sows exhibiting higher relative frequency of lying and lower frequency of standing posture (Enokida et al., 2011), spending less time feeding and drinking. The amount of feed intake during lactation influences subsequent reproductive performance, such as wean-to-first service interval and subsequent litter size (Koketsu et al., 1996). An inadequate nutrient and energy intake is expected to result in extended wean-to-oestrus interval with a lower percentage of sows in oestrus within seven days of weaning, reduced pregnancy rate and reduced embryo survival (Quesnel et al., 1998; Aherne et al., 1999). Especially protein restriction during lactation can have a negative impact on post weaning ovulation rate (Mejia-Guadarrama et al., 2002), whereas a crucial body protein mass loss can rapidly reduce ovarian function (Clowes et al., 2003) and increase the time required for expression of oestrus after weaning (King and Dunkin, 1986; Koketsu et al., 1996). In contrast to the above, we found that overgrown toes in front and rear feet were associated with more live-born and weaned piglets as well as with a shorter wean-to-first

service interval. The mean claw horn growth rate in sows is higher than the mean wear rate per month (6.3 mm vs 5.1 mm) (Amstel and Doherty, 2010); therefore, in higher parity sows, which are more prolific compared to younger ones (3–6 vs 1–2 parities) (Tantasuparuk et al., 2000), claw overgrowth may occur simply as a function of age.

The results of our study are limited to the extent that recording and scoring of lesions was based exclusively on farm personnel. Although there were training sessions for lesion characterization by the personnel, and the validity of a subsample of the recordings was verified by one of us, there were differences among herds. These differences were not only due to the unavoidable imperfect validity and repeatability of personnel scorings but also to the existing variations in management, productivity and genetic lines of sows. We opted not to analyze the associations within each herd but rather model the data obtained from all herds because the former approach would have reduced the statistical power to detect significant associations among individual or combinations of claw lesions with the investigated reproductive parameters. Our analytical approach was able to identify groups of closely related claw lesions among a larger set of fifty six variables describing lesions on feet of each sow, without losing any important information, and minimizing the possibility of finding associations 'due to chance alone' (Dohoo et al., 1997). We showed that sow reproduction ability was affected by different feet lesions, either having a combined effect on the examined reproductive parameters (factor score 10), or a discerned effect according to their location. Although generalization of these results is dangerous, since the data originated from three herds, when combined with those of other studies (Fitzgerald et al., 2012; Pluym et al., 2013), they point to the importance of

feet health and the need for general improvement in this area. Measures to improve feet health are expected to be of increased importance for both the longevity and productivity of sows in the modern swine industry, especially under the mandatory implementation of group housing (Anil et al., 2007; Fitzgerald et al., 2012) because claw lesions have a higher incidence of occurrence in loose compared to individually housed sows (Anil et al., 2007).

3.6. Conclusions

In conclusion, it is important to recognize that feet lesions, which are usually clinically unnoticed and therefore untreated, pose a constant challenge throughout a sow's life. Once a lesion is created the most probable outcome is its deterioration over time, thus explaining the overall negative impact on sow reproductive performance and longevity (Fitzgerald et al., 2012; Pluym et al., 2013). Claw lesions should not be regarded as independent injuries on specific sites, but rather as a complex, since they had a combined effect on the examined parameters. Claw lesions occur more frequently and severely in older sows (Dewey et al., 1993; Pluym et al., 2011; Fitzgerald et al., 2012), suggesting a time-related pattern of occurrence and likely a cumulative effect over time on the animal's reproductive performance.

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Chapter 4

Effect of diet supplementation with chelated zinc, copper and manganese on hoof lesions of loose housed sows³

M Lisgara, V Skampardonis, L Leontides

Department of Epidemiology, Biostatistics and Economics of Animal Production, School of Veterinary Medicine, University of Thessaly, 224 Trikalon st, 43100 Karditsa, Greece.

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4.1. Summary

Hoof lesions are very common among sows and have been associated with lameness, early removal and compromised welfare and productivity. Although housing conditions and management can have an external effect on hoof health status, the role of trace mineral intake is vital in developing hoof structure and integrity. Therefore, the objective of this study was to investigate the effect of a diet supplemented with organic complexes of trace minerals (Zn, Cu, Mn), partly substituting their inorganic form, on hoof lesions of sows in three Greek swine herds. A total of 518 sows were initially examined for hoof lesions and their respective severity was scored. For each hoof, the length of toes and dew claws were evaluated and five anatomical hoof sites, the heel, the sole, the white line, the wall and the coronary band, were examined for lesions. Subsequently, the same sows were re-scored after one or two gestations on diets supplemented with organic trace minerals, partly substituting their inorganic salt form (organic form of Zn 45 ppm, Cu 14 ppm and Mn 25 ppm of the total 125 ppm of Zn, 15 ppm of Cu and 40 ppm of Mn, respectively). The odds of the higher versus the lower lesion scores were significantly lower after than before the inclusion of the organic minerals in sows' diet, for each of the considered foot sites with the exception of the coronary band, with a distinct effect according to foot location. Specifically, on rear feet the improvement of hoof lesions was either smaller (for heel, sole and wall) than on front feet or not significant (for white line, toe and dew claw length). Additionally, for each foot site and herd examined, after the inclusion of the organic minerals, there were more sows with either the same or lower lesion score, with the exception of the toe and the dew claw length in one of

the herds. Within the specific conditions in the three studied herds, our findings highlight the role of chelated trace minerals in sows' hoof health,

suggesting an applicable and rewarding intervention to prevent hoof lesions.

Keywords: Sows, hoof lesions, organic minerals

4.2. Introduction

The reported frequency of occurrence of hoof lesions of sows in modern

swine herds is very high, usually exceeding 90 % of the studied sow

populations (Anil et al., 2007; Enokida et al., 2011; Lisgara et al., 2015). Hoof

lesions were associated with locomotor disorders (Anil et al., 2007; Lisgara et

al., 2015), early removal of sows (Anil et al., 2005; Jensen and Toft, 2009),

decreased litter weight, decreased number of liveborn piglets and increased

pre-weaning piglet mortality (Fitzgerald et al., 2012; Pluym et al., 2013a).

Thus, hoof lesions hamper the achievement of sows' optimum reproductive

performance. Moreover, hoof lesions and the associated lameness could

compromise sows' welfare, due to the feeling of pain and subsequent

locomotor impairment (Welfare Quality® Protocol, 2009). To promote sows'

welfare (Lynch et al., 2000) the European Council (Directive 2001/88/EC)

implemented the mandatory group housing of gestating sows in the European

Union, since January 2013. However, group housing in sows was associated

with increased leg and hoof disorders (Anil et al., 2005; 2007; Fitzgerald et al.,

2012). Despite the fact that housing conditions have an external effect on hoof

health status (Kroneman et al., 1993), the role of trace mineral intake is vital

for feet structure and integrity (Tomlinson et al., 2004). Keratinization of hoof

110

epidermis is controlled and moderated by a variety of bioactive molecules and hormones. Minerals (Zn, Cu, Mn, Se, Ca) and vitamins (A, D, E and biotin) have a substantial contribution in production and preservation of healthy keratinized tissues (Mülling et al., 1999; Van Riet et al., 2013). Particularly Zn, Cu and Mn were identified as key minerals in the process of keratinization (Mülling et al., 1999; Mülling et al., 2000) since they have an instrumental role in the activation of enzymes with catalytic, structural and regulatory function (Cousins, 1996; Van Riet et al., 2013).

Increasing the bioavailability of trace minerals improves their utilization and thus may help improve the integrity of keratinized tissues in cattle (Ballantine et al., 2002; Tomlinson et al., 2004) and swine (Anil, 2011). Many factors influence the bioavailability of trace minerals. The valence state of the mineral and its molecular form (inorganic or organic) in the diet are of major importance (Miles et al., 2000). These specific properties of the mineral may be responsible for the complexes they form with other components in the gut. which may either obstruct or facilitate the mucosal absorption, transport and/or metabolism of the mineral in target tissues (Miles et al., 2000). When conventional inorganic oxides and sulfates (e.g. ZnO, CuSO₄) in feed break down in the stomach, the released ions are free to interact with ligands, which will either allow them to remain soluble in the intestine or bind them to insoluble chelates, like phytate, and form low solubility salts which are unabsorbable (Peters, 2006). As a result, significant proportions of inorganic minerals are fecally excreted, stressing public concern about the environmental damage originated by intensive animal breeding operations (Gerber and Steinfeld, 2008). To protect livestock, consumers and the

environment the European Commission set maximum permitted levels for mineral concentrations in animal foodstuffs (Commission Regulation No 1881/2006).

Current hyper-prolific sow genotypes have greater mineral demands than older genotypes, because they farrow and lactate larger litters (Peters and Mahan, 2008; Peters et al., 2010). Thus, commercial sow diets are commonly formulated to exceed the NRC (1998) mineral requirements (Peters et al., 2010). Organic minerals, alternatively called chelated or proteinated minerals, are formed when the mineral is joined with an organic ligand, such as a protein or a specific amino acid (Carter, 1996). Organic minerals are more soluble and thus can be absorbed by the intestinal lumen more easily compared to inorganic minerals (Anil, 2011), utilizing intestinal uptake processes of amino acids (Power and Horgan, 2000). The biological advantage provided by organic compounds may be due to their unique coordination chemistry, which permits the formation of highly soluble. chemically stable products that resist digestion and interaction with antagonists in the gut (Brown and Zeringue, 1994; Yost et al., 2002). Therefore, using organic mineral sources instead of inorganic ones could help achieve a balance between animal trace mineral needs and their concentration in the diet, without affecting compliance with the limits of their inclusion rate in the feed (Poulsen and Carlson, 2008).

Therefore, the purpose of this study was to investigate the effect of diet supplementation with chelated zinc (Zn), copper (Cu) and manganese (Mn) on hoof lesions of sows in three Greek swine herds.

4.3. Materials and Methods

4.3.1. Study population

The study was carried out in three indoor, farrow-to-finish herds, with 330 (A), 160 (B) and 800 (C) sows, respectively, with Danbred (A, B) and Hermitage (C) genetics. For participation in the study the only criterion was the owners' written consent. Neither the health status of the sows' feet nor the frequency of locomotor disorders was considered for herd selection. During gestation sows were loose housed in static groups of eight to 12, with free-access to individual feed troughs in non-locking stalls, on combinations of concrete and slatted flooring. Pen design and flooring was in accordance to the requirements of the EU Directive for loose housing of gestating sows. All herds operated on weekly farrowing schedules.

4.3.2. Study design

At the beginning of the study, the hooves of the sows were examined for lesions by three farm employees, one on each herd, upon their entry to the lactation facilities. Training of the employees to identify, characterize and evaluate the severity of hoof lesions was done by two of the authors (ML and LL). Training involved an initial session at the clinics of the School of Veterinary Medicine, University of Thessaly (Karditsa, Greece) where the different anatomical sites of the hoof were identified and representative hoof lesions collected in the slaughterhouse were characterized and scored. Afterwards, training on scoring hoof lesions was repeated on each farm and employees were provided with a collection of pictures and the video of the training material, for referencing. Each sow's data were recorded on specially

developed paper data-capture forms. The primary author visited all farms once a month, collected the completed data-capture forms, and cross-checked the data by re-examining a random sample of 20 % of the sows with the responsible farm employee.

Until first scoring, sows were raised and fed throughout their life with diets containing only inorganic sources of minerals. The diets were in meal form and contained inorganic sources of Zn, Cu and Mn (125 mg/kg feed Zn from ZnO, 15 mg/kg Cu from CuSO₄ and 40 mg/kg Mn from MnO). Our nutritional intervention was the first occasion that the studied sows received a diet with an organic mineral source. Upon exit from the farrowing facilities the sows were offered gestation and later lactation diets in meal form supplemented with chelated trace minerals, from commercially available chelated mineral sources (Zinpro Performance Minerals®, Availa®Sow) as a partial substitution of their inorganic form (organic form of Zn 45 ppm, of Cu 14 ppm and of Mn 25 ppm of the total 125 ppm of Zn, 15 ppm of Cu and 40 ppm of Mn, respectively). From exit from the farrowing facilities until service, sows were offered a total of 4.0-4.5 kg daily of gestation feed. Thereafter, the daily amount of feed was 2.6-2.8 kg until 90 days and 3.2-3.5 kg from day 91 to 107 of gestation. The feed contained 12.6-12.8 MJ metabolizable energy (ME), 14.0-14.4 % crude protein, 6.2-6.4 g of digestible lysine, 15000 IU of vitamin A, 130 mg of vitamin E, 8.2-8.5 g of calcium (Ca), 5.2-5.5 g of phosphorus (P) and 0.45 mg of biotin per kg and was given either in one meal at 0700 h (Herd B and C) or split in half and offered in two meals at 0700 and 1600 h (Herd A). One week before the expected farrowing, sows were transferred to the lactation facilities where they were restricted fed until

five days after farrowing and then were offered ad libitum typical lactation diets containing 13.5 MJ ME/kg dry matter and 16.5-17.0 % crude protein, 7.5-7.7 g of digestible lysine, 17000 IU of vitamin A, 180 mg of vitamin E, 8.9-9.2 g of Ca, 5.7-6.0 g of P and 0.45 mg of biotin. The re-evaluation of the sows' hoof lesions was carried out after one or two gestations on diets supplemented with the organic trace minerals. For almost one third of the sows, re-scoring was performed at the first farrowing after the nutritional intervention, because they were not on farm for the second farrowing, whereas for the remaining sows re-scoring was conducted during the second farrowing after diet supplementation with chelated minerals.

The scoring system for hoof lesions applied in this study has been previously described in detail (Lisgara et al., 2015). Briefly, for each hoof, five anatomical sites were examined: the heel (soft keratinized epidermis on the ventral surface of the hoof towards the caudal end), the sole (hard keratinized epidermis cranial to the heel on the ventral surface of the hoof, including the junction between heel and sole), the white line (junction between sole and wall), the wall (hard keratinized epidermis on the dorsal surface of the hoof) and the coronary band. All hooves were examined for the presence of cracks, erosions, ulcers, bruises, separation along the white line and hyper-keratinization and the respective anatomical sites were scored in a severity scale ranging from 0 to 2, where score 0 was given to hoof sites with no lesions or very small superficial ones. For the sole, score 1 was given to hooves with erosions and/or superficial heel-sole separation and 2 to hooves with ulcers and/or severe heel-sole separation. For the heel, score 1 was given to hooves with hyper-keratinization and erosions and 2 to hooves with

hyper-keratinization and ulcers. For the white line score 1 was assigned to hooves with superficial separation and 2 to hooves with deep separation. For the wall, the score was 1 when cracks often accompanied by bruises were observed and 2 when deep cracks were noted. For toe and dew claw length, score 0 was assigned to toes and dew claws with normal length, score 1 to extended toes and dew claws touching the floor when the animal was standing and score 2 to overgrown and twisted or cracked toes and overgrown and twisted or crushed dew claws. For the coronary band the score ranged from 0 to 1 (0 = no lesions, 1 = lesions of any kind, edema, hemorrhage and/or necrosis).

4.3.3. Statistical analysis

All statistical analyses were performed using Stata 13.1 (Stata Statistical Software. College Station, TX) and evaluated for significance at the 5 % level. The total score for the four feet for each anatomical site considered was obtained by adding the respective scores of each of the five hoof sites, toes and dew claws. Therefore, for all anatomical sites except the coronary band, the total score for the four feet could range from 0 to 16; for the coronary band, the total score varied between 0 and 8. Comparison of the proportion of sows with the same or lower total lesion score for each hoof site with the proportion of sows with higher score, before and after one or two gestations on diets supplemented with the organic trace minerals was performed by Mc Nemar's χ^2 test for symmetry.

The possible effect of the dietary intervention with organic trace minerals on the severity of lesions on each hoof site considered, was

estimated in seven mixed-effect, either ordinal (for all hoof sites except for the coronary band) or binary (for the coronary band), logistic regression models in GLLAMM (Rabe-Hesketh et al., 2005). These models included the score of the anatomical site considered as the dependent variable and the dietary status (before or after the supplementation of the diet with organic trace minerals), the foot (front or rear), the toe (medial or lateral), the sow's parity and the farm of sow's origin as the independent variables. The latter two variables were forced in the models in order to control for their likely confounding effects. Furthermore, these models included a random-effect term for sow, a random-effect term for foot nested within sow and a randomeffect term for toe nested within foot in order to account for the hierarchical structure of multiple measurements and repeated scoring on the same animal, foot and toe. The proportional odds assumption for the fitted ordinal regression models was verified graphically, by assessing the plots of the resulting empirical cumulative logit functions of each of the dependent variables considered. After initial fit of the models to the data all possible twoway interactions between the fixed effects for diet, foot and toe were created, were then offered one-by-one to the initial models and evaluated for significance at the 5 % level. The final models contained the fixed-effects for diet, foot and toe, the significant two-way interactions between the fixed effects and the random-effects.

4.4. Results

A total of 518 sows, of which 124 were in herd A, 112 in herd B and 282 in herd C, were initially scored and subsequently re-scored after one (186/518,

35.91 %) or two (332/518, 64.09 %) gestations on diets supplemented with chelated trace minerals, as a partial substitution of their inorganic form. Upon initiation of the study, the median (range) parity of the studied sows was 1(1-8), 3(1-9) and 3 (1-6) for herd A, B and C, respectively. The proportion of sows with at least one lesion on any foot, at first scoring, was 120/124 (96.77 %), 110/112 (98.21 %) and 256/282 (90.78 %) in herds A, B and C, respectively. At second scoring 118/124 (95.16 %), 106/112 (94.64 %) and 250/282 (88.65 %) of sows in herds A, B and C, respectively, had at least one hoof lesion. By herd, in decreasing order, the four most frequently recorded lesions at first scoring were: In herd A, 112/124 (90.32 %) sows had at least one overgrown toe and an equal proportion of at least one overgrown dew claw [median of total lesion score 4 (range 0-8) and 3 (0-15), respectively], 56/124 (45.16 %) [median of total score 0 (range 0-8)] had at least one heel lesion and 50/124 (40.32 %) [median of total score 0 (range 0-7)] at least one wall lesion. In herd B, 100/112 (89.29 %) [median of total score 2 (range 0-10)] and 82/112 (73.21 %) [median of total score 1 (range 0-4)] sows had at least one heel or one wall lesion, respectively, while those with at least one overgrown toe or dew claw were, correspondingly, 80/112 (71.43 %) [median of total score 2 (range 0-7)] and 76/112 (67.86 %) [median of total score 2 (range 0-7)]. In herd C, sows with at least one heel lesion or one overgrown dew claw were 228/282 (80.85 %) [median of total score 4 (range 0-16)] and 198/282 (70.21 %) [median of total score 2 (range 0-16)], respectively, while those with at least one wall or one sole lesion were 120/282 (42.55 %) [median of total score 0 (range 0-8)] and 114/282 (40.43 %) [median of total score 0 (range 0-8)], respectively.

In Figures 4.1, 4.2 and 4.3, the proportion of sows with improvement or no change in the severity (total score) of lesions on the different foot sites versus the proportion of those with lesion deterioration, after diet supplementation with organic trace minerals are presented by herd. In herd A, the proportion of sows showing improvement or no change in the severity of lesions on the sole, the heel, the white line, the wall and the coronary band, was higher (P < 0.05) than the one showing deterioration, whereas there was no difference (P = 0.72) for toe length. In contrast, for dew claw length, the proportion of sows with the same or lower score was lower (P < 0.001) than the one with higher score. In herds B and C, the frequency of improvement or no change in the total lesions score for all anatomical sites of the feet was higher (P < 0.001) compared to the frequency of deterioration; the only exception was the heel lesions of sows in herd B, where the before and after frequencies marginally did not differ (P = 0.06).

Figure 4.1. Frequency of improvement or no change and worsening in the severity of hoof lesions after diet supplementation with organic minerals, in herd A. The proportion of sows (n = 124) showing improvement or no change and worsening in the severity of lesions on SL (sole), HL (heel), WL (white line), WA (wall), CB (coronary band), TL (toe length) and DCL (dew claw length), after one or two gestations on a diet with organic trace minerals (Zn, Cu, Mn). Mc Nemar's χ^2 test for symmetry was used to detect differences between the proportion of sows showing improvement or no change and the sows showing worsening in the severity of the hoof lesions, *P<0.05

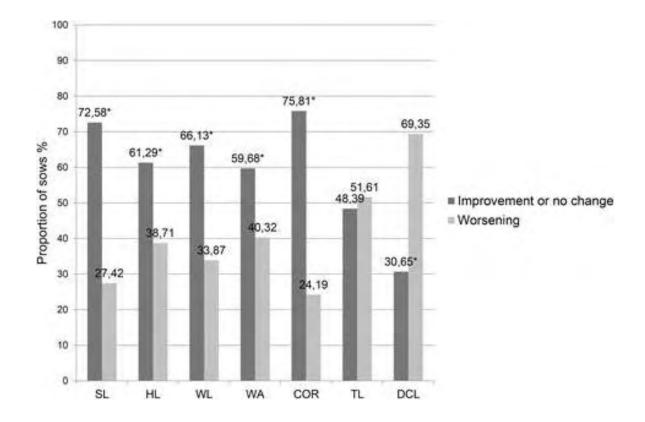


Figure 4.2. Frequency of improvement or no change and worsening in the severity of hoof lesions after diet supplementation with organic minerals, in herd B. The proportion of sows (n = 112) showing improvement or no change and worsening in the severity of lesions on SL (sole), HL (heel), WL (white line), WA (wall), CB (coronary band), TL (toe length) and DCL (dew claw length), after one or two gestations on a diet with organic trace minerals (Zn, Cu, Mn). Mc Nemar's χ^2 test for symmetry was used to detect differences between the proportion of sows showing improvement or no change and the sows showing worsening in the severity of the hoof lesions, *P<0.05

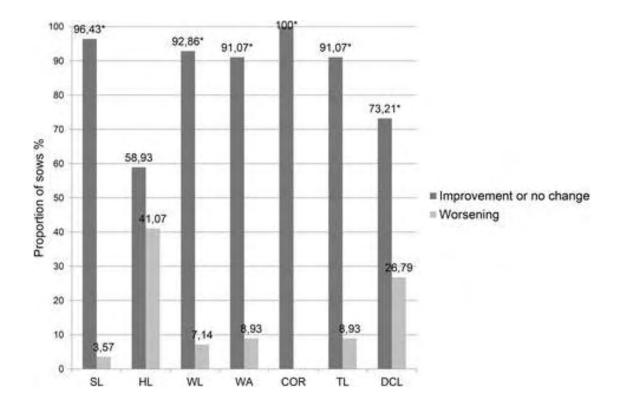
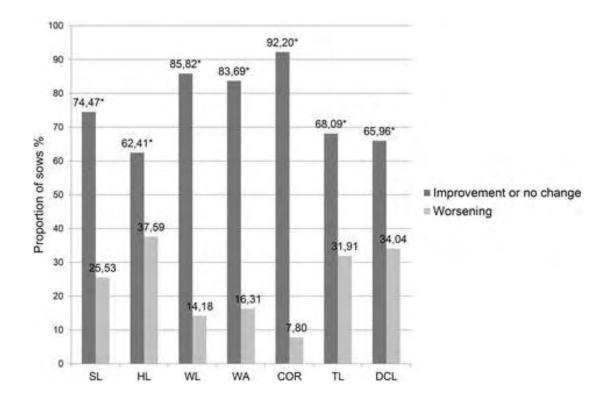


Figure 4.3. Frequency of improvement or no change and worsening in the severity of hoof lesions after diet supplementation with organic minerals, in herd C. The proportion of sows (n = 282) showing improvement or no change and worsening in the severity of lesions on SL (sole), HL (heel), WL (white line), WA (wall), CB (coronary band), TL (toe length) and DCL (dew claw length), after one or two gestations on a diet with organic trace minerals (Zn, Cu, Mn). Mc Nemar's χ^2 test for symmetry was used to detect differences between the proportion of sows showing improvement or no change and the sows showing worsening in the severity of the hoof lesions, *P<0.05



With the exception of the coronary band, final models for all other foot sites considered included an interaction between the dietary status and the foot. In addition, the final models for the severity of heel lesions and toe length included an interaction between the dietary status and the toe. In Table 4, the odds ratio for higher versus lower lesion severity score(s) (lesion score 2 vs score 1 or 0 and lesion score 2 or 1 vs score 0) for each foot site, are presented. Specifically, after the inclusion of the organic minerals in the diets, the odds of sole lesion score 2 versus 1 or 0 and score 2 or 1 versus 0 (higher versus lower score(s)), were 0.28 (95 % CI: 0.21, 0.38) and 0.48 (0.37, 0.61) times lower, for front and rear feet respectively, than before the inclusion of the organic minerals. Similarly, the odds of higher versus lower heel lesion score(s), after one or two gestations on diets with organic minerals, were 0.46- (95 % CI: 0.37, 0.57) and 0.69- (95 % CI: 0.55, 0.82) fold lower, for front and rear feet, respectively, and 0.34 (95 % CI: 0.27, 0.44) and 0.46 (95 % CI:

0.37, 0.57) times lower, for medial and lateral toes respectively, than before the inclusion of the organic minerals. The odds of higher versus lower white line lesion score(s) on sows' front feet were 0.49 (95 % CI: 0.35, 0.69) times lower, after than before the inclusion of chelated minerals in the diets. However, there was no difference (P = 0.13) in the odds of higher versus lower score(s) of white line lesions on rear feet. For wall lesions, the odds of higher versus lower score(s) were 0.32 (95 % CI: 0.25, 0.42) and 0.50 (95 % CI: 0.40, 0.66) times lower on front and rear feet, respectively, after the inclusion of the organic minerals. For toe length, the odds of higher versus lower score(s) were 0.26-fold (95 % CI: 0.19, 0.35) lower for front feet and 0.36 (95 % CI: 0.28, 0.51) and 0.26 (95 % CI: 0.19, 0.35) times lower for medial and lateral toes respectively, after than before diet supplementation with organic mineral sources. We found no difference (P = 0.35) in the toe length score on rear feet before and after the inclusion of chelated minerals in sow diets. For the dew claw length the odds of higher versus lower score(s) after the inclusion of the organic minerals in the diets were 0.57 (95 % CI: 0.46, 0.70) times lower for front feet but there was no difference (P = 0.21) in the odds of dew claw length of rear feet. Lastly, there was no significant (P=0.17) effect of the chelated mineral diets on the likelihood of lesions on the coronary band. For all hoof sites examined there was a significant (P < 0.05) association between the severity of hoof lesions and sows' parity. Specifically, the odds of sole lesion score 2 versus 1 or 0 and score 2 or 1 versus 0 (higher versus lower score(s)), were 1.23 (95 % CI: 1.14, 1.33) times higher for one unit increase of sows' parity. The odds of higher versus lower lesion score(s) were, 1.08 (95 % CI: 1.02, 1.15) and 1.12 (95 % CI: 1.02,

1.23) times higher for one unit increase of sows' parity, for heel and white line lesions respectively. Similarly, for lesions on the wall and the coronary band the odds of higher versus lower lesion score(s) were 1.18 (95 % CI: 1.12, 1.27) and 1.16 (95 % CI: 1.03, 1.31) times higher, respectively, for one unit increase of sows' parity. For toe and dew claw length, the odds of higher versus lower lesion score(s) were 1.15- (95 % CI: 1.06, 1.23) and 1.08- (95 % CI: 1.01, 1.16) fold higher, respectively, for one unit increase of sows' parity.

Table 4. Associations between the severity of hoof lesions and the dietary status of the sows

Foot	Foot	Odds ratio ^a	95 % CI	Toe	Odds ratio ^a	95 % CI
site	location			location		
SL	Front	0.28*	0.21, 0.38	NA	-	-
	Rear	0.48*	0.37, 0.61		-	-
HL	Front	0.46*	0.37, 0.57	Medial	0.34*	0.27, 0.44
	Rear	0.69*	0.55, 0.82	Lateral	0.46*	0.37, 0.57
WL	Front	0.49*	0.35, 0.69	NA	-	-
	Rear	0.78	0.57, 1.07		-	-
WA	Front	0.32*	0.25, 0.42	NA	-	-
	Rear	0.50*	0.40, 0.66		-	-
TL	Front	0.26*	0.19, 0.35	Medial	0.36*	0.28, 0.51

	Rear	0.88	0.70, 1.13	Lateral	0.26*	0.19, 0.35
DCL	Front	0.57*	0.46, 0.70	NA	-	-
	Rear	0.88	0.74, 1.07		-	-

SL sole, HL heel, WL white line, WA wall, TL toe length, DCL dew claw length Mixed-effect ordinal logistic regression models associating the severity of lesions (scaled from 0 to 2) on six foot sites of 518 sows, with a diet supplemented with organic trace minerals over a period of one or two gestations (After), following a conventional inorganic mineral source diet (Before), in terms of its interaction with the location of the foot (front or rear) or the location of the toe (lateral or medial) a Odds ratios (After/Before) were adjusted for foot location (front or rear), toe location (lateral or medial), sows' parity and farm NA not applicable; the effect of the applied diet was not associated with toe location $^*P < 0.05$

4.5. Discussion

Previous studies have reported on the effect of flooring and management on the frequency and severity of hoof lesions of sows (Vestergaard et al., 2008; Spoolder et al., 2009; Pluym et al., 2013b). There are, however, sparse reports on the effect of nutrition on sow hoof lesions (Anil et al., 2009; 2010; Anil, 2011), with the exception of biotin supplementation (Greer et al., 1991; Watkins et al., 1991). In studies conducted in dairy cows, replacing sulfate minerals with an organic source, decreased severity of white line lesions, sole ulcers and heel erosions (Ballantine et al., 2002; Nocek et al., 2006; Siciliano-Jones et al., 2008). Additionally, it was shown that the supply of a combination of complexed trace minerals is more beneficial to claw integrity than supplying a sole complexed trace mineral due to synergistic effects (Nocek et al., 2000). In this longitudinal study, we investigated the effect of gestation and lactation

sow diets supplementation with organic complexes of trace minerals, over one or two gestations, on hoof lesions of sows, in three Greek swine herds with typical, at least to nationally applied standards, general management and housing. The average claw horn growth rate of sows was recently estimated at about 6.3 mm per month, whereas over the same period the overall mean wear rate was about 5.1 mm (Van Amstel and Doherty, 2010). The average claw length of sows is about 5 cm (Van Amstel and Doherty, 2010). Thus, a period of a reproductive cycle including one full-term gestation and four weeks lactation may be considered minimum for complete hoof horn regeneration.

Trace minerals have an important role in maintaining horn integrity (Tomlinson et al., 2004; Van Riet et al., 2013). The use of chelated minerals could enhance their absorption and utilization in the body (Power and Horgan, 2000; Peters, 2006), probably due to increased bioavailability (Matsui et al., 1996; Veum et al., 2004), and thus the integrity of keratinized tissues (Ballantine et al., 2002; Anil, 2011). More specifically, the benefit to hoof keratinization of adequate Zn, Cu and Mn supplementation is related with the reinforcement of the epidermal junctions with the corium, making them less susceptible to separation or breakdown, and ultimately the improved integrity of the epithelial tissue (Nocek et al., 2000). In general, our results suggest that chelated minerals have a beneficial effect on the health status of the majority of hoof anatomical sites and the severity of lesions. However, there were hoof sites on the rear feet which had no decrease in the risk of lesion severity.

The severity of hoof lesions is expected to increase by increasing sow parity (Lisgara et al., 2015); this "parity-effect" was accounted for in our analysis. Therefore, interventions (managerial or nutritional) which result to a

relatively stable or unaffected status of hoof lesions while sows carry out more gestations can be considered to have a positive effect (Anil, 2011). We found that after inclusion of the chelated minerals, lesions on the heel, the sole and the hoof wall had decreased severity which was more pronounced on front than rear feet and on medial than lateral heel bulbs. It has been reported that lesions on these sites are more frequent and severe on rear than on front feet (Gjein and Larssen, 1995; Enokida et al., 2011; Lisgara et al., 2015). Moreover, the lateral heel bulb usually carries most of the sow's weight and thus it is more prone to injuries compared to the medial heel bulb (Webb et al., 1984; Anil et al., 2007; Pluym et al., 2011). The white line is the junction of wall and sole horn. Lesions on this site, which were reported to be more frequent and severe on rear compared to front feet (Gjein and Larssen, 1995), corresponded to deep separation of the wall and sole junction. This separation may facilitate the invasion of bacteria into the corium, resulting in infection and inflammation. Therefore, since the reduction of the severity of sole and hoof wall lesions was slower on rear that front legs, our failure to demonstrate significant improvement on white line lesions may indicate that on rear feet, the diets supplemented with chelated trace minerals, may probably not avert, at least at the applied inclusion rate and/or time interval, the course or deterioration of such severe lesions. We have recently shown that sow laminitis may frequently occur (Varagka et al., 2016). We histologically examined claws of culled sows and recorded histological changes (e.g. hyperplasia, edema, hyperemia, hemorrhage, white blood cells), that had been previously described in cases of equine and bovine laminitis (i.e. inflammation of the lamellar corium) (Obel, 1948; MacLean,

1971; Wattle, 2000; Karikoski et al., 2015). An inflammation in the corium may lead to interference in the supply of nutrients (Hoblet and Weiss, 2001), since keratinocytes are dependent on receiving oxygen and nutrients from the fine microvasculature of the corium by diffusion across the basement membrane (Mülling, 1999). Moreover, we found a positive association between the existence of these pathological changes and higher total lesion score of the claw. These changes were more frequently observed on the rear feet and on the lateral toes where, we and others also found that hoof lesions are more frequent and severe (Anil et al., 2007; Enokida et al., 2011; Lisgara et al., 2015). Therefore, for hooves with more severe lesions (higher lesion score), which are more likely on rear feet and on lateral toes, there would be a greater interference in the supply of nutrients and, thus, their improvement may be a more demanding and/or time consuming process compared to the improvement of the respective lesion scores on front feet and on medial heel bulbs. Thus, for decrease in the risk of lesion severity on these sites, either the use of chelated minerals in the sow diets should be longer than the period investigated or these lesions on these sites are not reversible with the inclusion rate of chelated minerals investigated in this study. Lastly, the fact that the chelated minerals supplemented diets had no effect on the occurrence of lesions on the coronary band was most likely attributed to the very low frequency of lesions on this site (2.2 %, only 182 affected hooves in the totally 8288 examined, before and after the inclusion of chelated minerals in sows' diet).

Overgrown toes and dew claws are two of the most frequently occurring hoof lesions (Lisgara et al., 2015). When gilts are developing to

sows, their legs tend to conform to the shape and size of toes, which is defined by the combined effect of genetic, managerial and nutritional factors. Thus, excessive toe length in sows may occur when gilts are not selected for proper feet development (Stalder et al., 2010). Because claw horn growth rate is greater than the respective wear rate, toe overgrowth could occur as a function of age, especially when sows are not provided with enough space for adequate exercise (Van Amstel and Doherty, 2010). An older study suggested that routine hoof trimming may be a preventive measure against the development of hoof lesions (Kroneman et al., 1992). More recent ones, did not recommend routine trimming for lesion reduction because they measured no clear effect on hoof lesion development and sow longevity; therefore the additional labor and costs associated with regular claw trimming were not justified (Vestergaard et al., 2008; Spoolder et al., 2009). However, these studies did not combine nutritional interventions, such as inclusion of chelated Zn, Cu and Mn, with trimming. It may be that the use of organic minerals in sow diets prevents the development of hoof lesions on the heel, the sole and the hoof wall but offers less in the prevention of toe overgrowth, which may be affected by a variety of factors (i.e. genetic, housing, flooring, age) (Newman et al., 2015). The length of dew claws, on the other hand, is also affected by genetic factors and age but not by housing and flooring because they do not normally touch the ground or bear any weight. In relevant reviews (Tomlinson et al., 2004; Van Riet et al., 2013) and published studies (Anil, 2011) no effect of trace minerals on dew claw overgrowth was suggested or found. Therefore, we also expected limited effect, if any, of the nutritional intervention on dew claws overgrowth. We have recently demonstrated that all hoof lesions,

including overgrowths, act synergistically to impair the sow's locomotor ability (Lisgara et al., 2015). Therefore, in an attempt to collectively interpret the results of the present and previous studies we advocate routine trimming of overgrown toes (Newman et al., 2015) and dew claws, in herds where these lesions are frequent, in addition to proper gilt selection and use of organic minerals in sow diets for mitigation of the risk of sow feet problems.

The results of our study are limited to the extent that recording and scoring of lesions was conducted by farm personnel. Although there were training sessions for lesion characterization by the personnel, and the validity of a subsample of the recordings was verified by one of us (ML), there were differences among herds. These differences were due not only to the unavoidable imperfect validity and repeatability of personnel scorings, but also to the existing variations in management, productivity, and genetic lines of sows. Moreover, since data of our study originated from three herds, the generalization of our results to the national Greek sow herd is dangerous. However, the longitudinal nature of our study provided, as a within-subject analysis, an internal validity independent of random assignment and a substantial boost in statistical power (Charness et al., 2012). Besides the high statistical efficiency, the employed intra-subject comparisons, with each sow acting as its own control, controlled for the influence of all possible confounders, genetic factors or traits that remained constant over time for each sow, automatically by design (Maclure, 1991; Diggle et al., 1994; Navidi, 1998).

4.6. Conclusions

Within the specific conditions in the three studied herds, our findings highlight the role of chelated trace minerals in sows' hoof health, suggesting an applicable and rewarding intervention to prevent hoof lesions.

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Chapter 5

5. General discussion and future perspectives

The development of this thesis was motivated by the growing concern of Greek pig producers about the likely negative effects, after the implementation of the EU Directive 2001/88/EC for mandatory group housing of pregnant sows, on sow hoof health, longevity and overall productivity. The thesis was part of a larger project, which aimed at assisting the implementation and full compliance of the Greek swine industry with the EU Directive by (1) increasing our understanding about claw lesions as a risk factor for sow longevity and welfare in Greek swine herds, and by (2) preventing, through scientifically proven nutritional adjustments, reduced sow longevity and productivity in the Greek swine herds. Furthermore, the project also aimed at broadening the pig farmers' options of using low-cost agricultural by-products in the preparation of fibrous rations for dry sows and growing maiden gilts, an additional requirement of the Directive.

For serving the project goals and the thesis objectives, we initially formulated and documented a claw lesion detection and scoring system for each of the five anatomical sites of the claw, namely the sole, the heel, the white line, the wall, and the coronary band. Additionally, we also considered and scored toe and dew claw overgrowth. By applying the scoring system, we found very high frequency and severity of sow claw lesions, especially on the heel, and very frequently overgrown toes and dew claws in the studied herds. Lesions that were located on the white line, the sole, the wall, and the overgrown toes and dew claws, were associated with lameness. Interestingly, the more severe the claw lesions the higher the lameness scores were.

scores. Individual lesions had a discerned effect on lameness which depended on their location. The simultaneous presence of lesions on more than one-foot site, on the same or a different foot, had a multiplicative effect on the likelihood of lameness.

Sows' reproductive performance, expressed in terms of the number of liveborn piglets, the number of weaned piglets, and the wean-to-first service interval was affected by several claw lesions. Similarly to findings for lameness, sows' reproduction ability was affected by different feet lesions, either having a combined effect on the examined reproductive parameters, or having a discerned effect according to their location. The number of live-born piglets was negatively associated with lesions on heel and sole of front feet.

The number of weaned piglets was also negatively associated with lesions on heel of any foot, on sole of front feet and on white line, sole and wall of rear feet, while the wean-to-first service interval was positively associated with lesions on heel of any foot, on sole of front feet and of dew claw length of front feet.

We found that the inclusion of chelated zinc, copper and manganese, for partial substitution of their inorganic form in sows' diets significantly improved the general health of the claws. Comparison of the sow claws (paired observations on the same animal) revealed a higher likelihood for lower lesion scores after than before the inclusion of the organic minerals, on all of the considered foot sites but the coronary band. Improvement was more pronounced on front than on rear feet and on medial than on lateral heel bulbs. The frequency and severity of overgrown claws and dew claws on rear lateral toes were not improved after the organic mineral supplementation.

Our clinical observations presented in this thesis, were supported by histopathologic analyses conducted in other studies which were also supported by the same project. In those studies it was found that histologic changes such as lamellar hyperplasia, presence of white blood cells, hyperemia, hemorrhage, edema, and necrosis of the dermis, which were present in cases of bovine and equine laminitis were also observed in the dermis and the epidermis of the sows' claws. Supplementation of sow diets with the chelated minerals reduced the incidence of histologic changes and improved the appearance of the horn architecture, manifested especially in the form of smaller tubules and increased density of horn tubules. Increased diameter of the horn tubules was associated with genesis of qualitatively inferior horn and increased tubular density was associated with enhanced hoof hardness. The inflammatory changes recorded likely obstruct the oxygen and nutrient supply of the corium, where epithelial cells are transformed to keratinocytes, compromise the quality of the hoof horn, and ultimately lead to production of low-quality hoof horn which is vulnerable to mechanical stresses, injuries and ascending infections.

The generally reported high prevalence of claw lesions in modern sows, their multifactorial etiology, and the fact that treatment of infected claw lesions by drug administration (local and systemic) is frequently unrewarding, points to the direction of prevention through changes in husbandry, housing and nutrition. When the results of this thesis are seen together with those of the studies documenting improvement of histologic changes in the corium of the claws and the morphometry of the hoof horn, they offer a reasonable explanation of the mode of action of mixtures of chelated zinc, copper and

manganese. Usually, pig farmers tend to perceive that sow nutrition is more easily amenable to changes than general sow housing conditions. However, nutrition should not be considered a panacea. In herds with high incidence of hoof related locomotor disorders in sows, improvements in housing, flooring and general sow husbandry, should always, in medium-term at least, complement nutritional changes.

Noteworthy, the incidence of overgrown toes and dew claws, which was found very high in the studied populations and complicated the severity of sow lameness, was not reduced after using the chelated mineral nutrition. In recent studies it was reported that overgrown toes and dew claws were found not to be associated with laminitic changes in the claws. Overgrown toes and dew claws increase the risk for trauma to the claws, may alter the animals' posture and gait and reduce its general locomotor ability. Because routine onfarm claw trimming has been recently found not to improve sow retention and was a measure with sub-optimal return-on-required investment, further research is needed to investigate the predisposing factors for toe and dew claw overgrowth and to generate a more holistic preventive strategy against claw overgrowth.

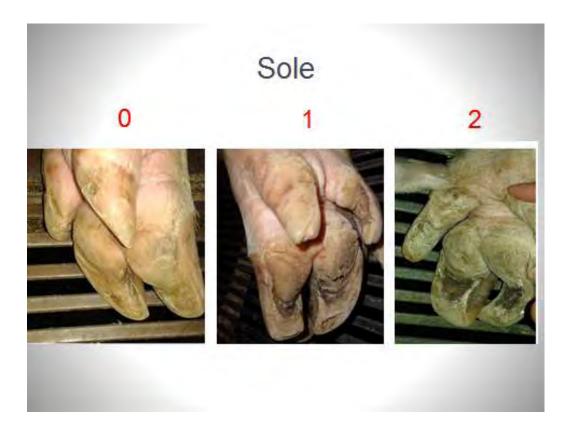
It is generally accepted that claw lesions may affect the postural behavior of the sow, with affected sows exhibiting higher relative frequency of lying and lower frequency of standing posture, spending less time feeding and drinking. Prolonged lying on floors covered with feces or hardly ever disinfected (such as the floors of continuously occupied dry sow stables) in combination with reduced water intake could predispose to infection of the urinary and/or genital tract. These infections can lead to reproductive failure,

perinatal diseases or sudden death. Indeed, urinary tract infections (UTI) leading to the cystitis and pyelonephritis syndrome are considered for many years as the leading infectious cause for sow mortality. Yet, the associations between sow hoof health and UTI have not been elucidated, although the latter are considered among the most important infectious causes of suboptimal reproductive performance, leading to early sow removal.

Appendix

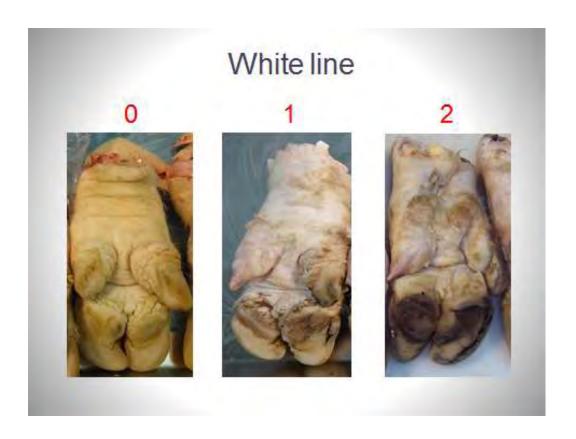
I. A photographic documentation of the scoring system applied for the evaluation of lesions on seven sites of the sow's foot.

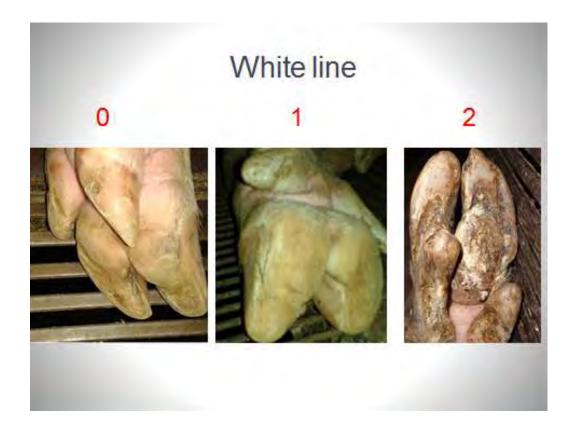


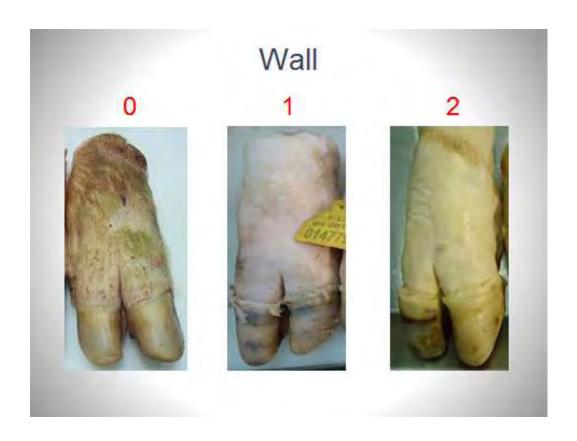






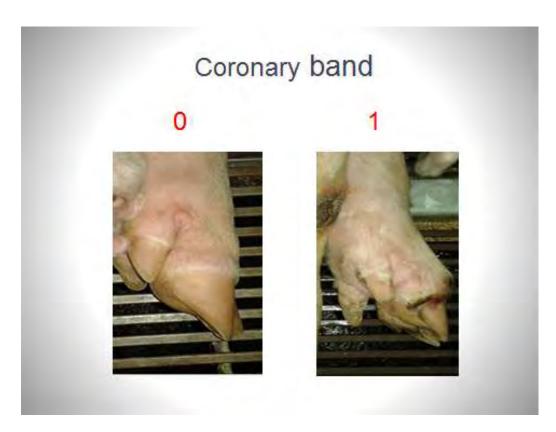


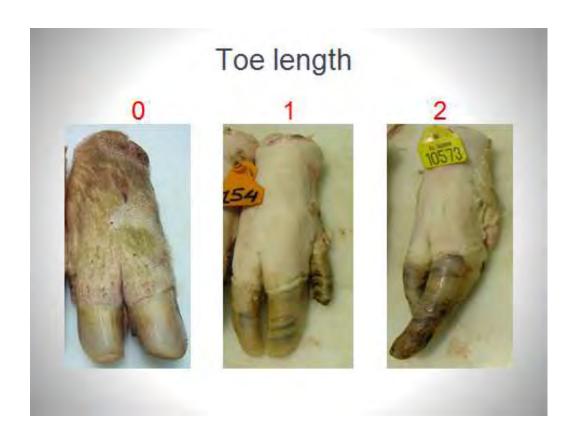


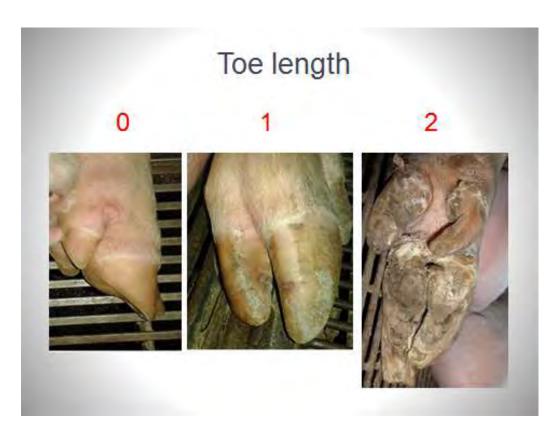


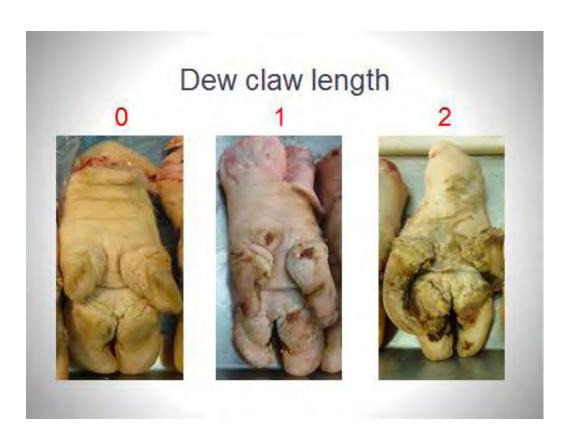














II. List of publications from the present thesis

Parts of the present thesis have been presented in national and international Symposia and have been published in national and international peer-reviewed journals. The list of those publications, in chronological order, follows:

- Lisgara M, Kostoulas P, Skampardonis V, Leontides L. Claw lesions in individually and group housed sows in Greek swine farms. *Proceedings* of the 23rd International Pig Veterinary Society Congress. Cancun, Mexico, 8-11 June 2014, p. 131.
- Lisgara M, Skampardonis V, Kouroupides S, Leontides L. Clinical evaluation of feet lesions and associated lameness of sows in three Greek swine herds. *Proceedings of the 13th National Conference of* Veterinary Medicine. Athens, Greece, 8-10 May 2015, p. 61.
- Lisgara M, Skampardonis V, Kouroupides S, Leontides L. The effect of chelated manganese, copper and zinc on feet lesions of sows in three Greek swine herds. *Proceedings of the 13th National Conference of* Veterinary Medicine. Athens, Greece, 8-10 May 2015, p. 60.
- 4. Lisgara M, Skampardonis V, Kouroupides S, Leontides L. Hoof lesions and lameness in sows of three Greek swine herds. *Journal of Swine Health and Production*. 2015;23:244–251.

- Lisgara M, Skampardonis V, Angelidou E, Kouroupides S, Leontides L.
 Associations between claw lesions and reproductive performance of sows in three Greek herds. *Veterinarni Medicina*. 2015;60(8):415–422.
- Lisgara M, Skampardonis V, Leontides L. Effect of diet supplementation with chelated zinc, copper and manganese on hoof lesions of loose housed sows. *Porcine Health Management*. 2016;2:6.
- Lisgara M, Skampardonis V, Angelidou E, Kouroupides S, Leontides L.
 Associations between hoof lesions and reproductive performance of sows in three Greek herds. *Proceedings of the 24th International Pig Veterinary Society Congress*. Dublin, Ireland, 7-10 June 2016, p. 120.