

Trends in premature death in pet rabbits.

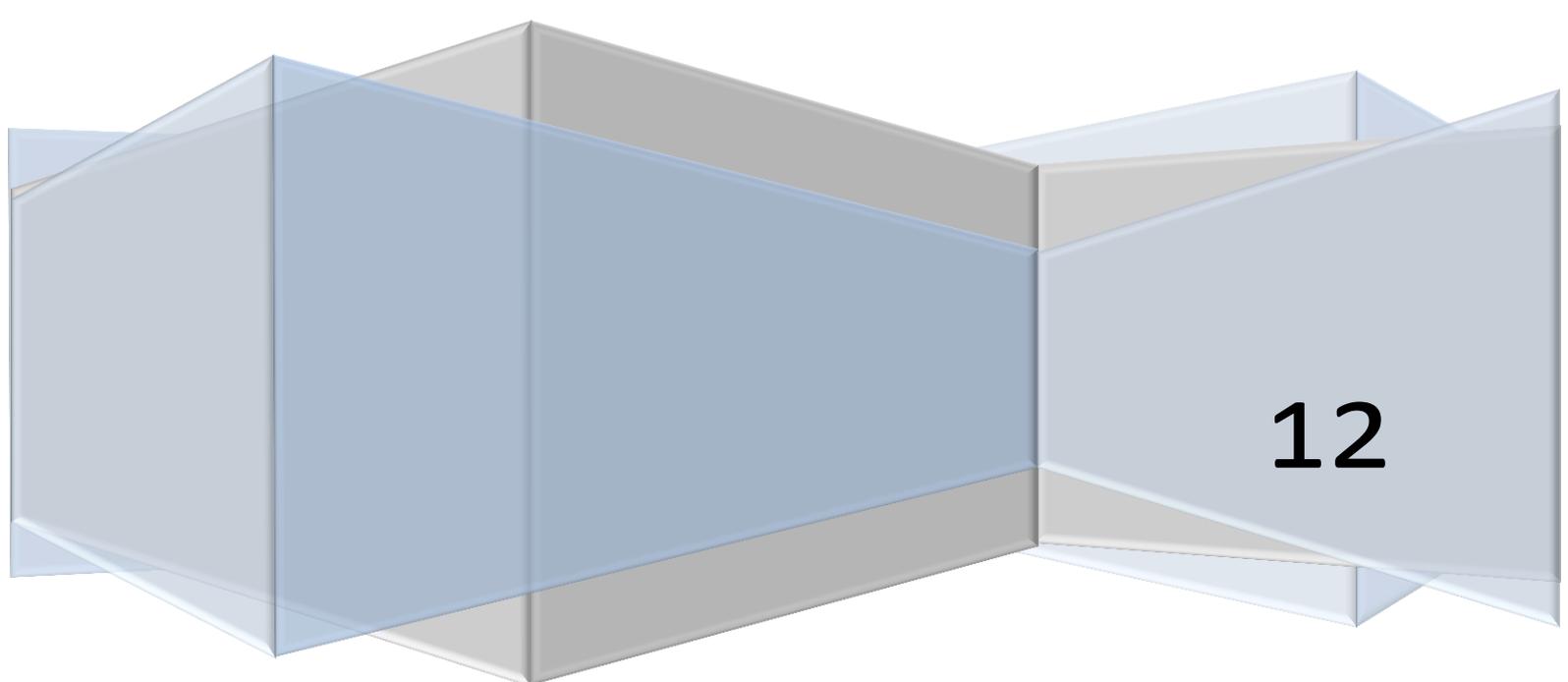
Welfare research small mammals.

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Abstract:

A pet rabbit can reach the age between 8 and 12 years, when given the proper care [5,6,13,17]. In the Netherlands, the average age of pet rabbits is between 3 and 4.5 ($\pm 0,3$) years of age [4,5]. The Veterinary Pathology Diagnostic Centre (VPDC) of the Veterinary Medicine Faculty at Utrecht University started a 3 year lasting research on 1 October 2009, the “Welfare research small mammals”. To investigate the causes of death in young pet rabbits and evaluates options to expand the average age of rabbits, guinea pigs, rats and ferrets. This paper evaluates the results of the rabbits submitted to this project from November 2010 to February 2011. 38 young deceased rabbits between the ages 2 to 36 months old were submitted for necropsy. The owners of the deceased rabbit and the treating veterinarian provided zootechnical information, which was compared with the results of the pathological results. 11 hypotheses were answered. Results show that most rabbits died of an infectious agent and that none of the rabbits which were vaccinated against VHD and myxomatosis died of these diseases. There is also evidence that there is a relationship between housing rabbits individually and lung infections, the number 1 cause of death in this study. And that the pathological diagnosis didn't correspond with the veterinarian diagnosis. In this study, results show that there are more factors responsible for the early death of pet rabbits. Recommendations for the owners of pet rabbits and for veterinarians who work with rabbits were made based on the results of this research and on the literature study.

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Introduction:

The rabbit is a popular pet in the Netherlands, there are almost a million (0,98 million) pet rabbits in the Netherlands [5,6]. The rabbit is now the third most popular pet in the Netherlands [5]. The life expectancy of a rabbit is 8 to 12 years [1, 4, 26]. Studies show that the average age in the Netherlands is far beneath this number. The study of Caneel 2000, showed that the average age is 4,5 ($\pm 0,3$) years old, and the research of Leenstra 2009 had a result of only 3 years of average age[27].

The Veterinary Pathology Diagnostic Centre (VPDC) of the Veterinary Medicine Faculty at Utrecht University started a 3 year lasting research on 1 October 2009, the “Welfare research small mammals”. This research is funded by the Dutch Ministry of Economics, Agriculture and Innovation. The aim is to investigate the cause of death in early deceased pet rabbits, guinea pigs, ferrets (2-36 months old) and rats (2-12 months old). This knowledge is the basis to provide welfare advice to prevent early death in these animals. Owners of young predeceased rabbits, guinea pigs, ferrets and rats can free of charge, participate their animal in this research. When the research started, the age limits to participate in the research were set at 6-36 months old for rabbits, guinea pigs and ferrets, and for rats 4-12 months. But after one year of research, it was decided that rabbits, guinea pigs and ferrets should be between 2-36 months, and rats between 2-12 months old to participate. The owner provides extensive zootechnical information, to get an insight of the health and welfare of this animal (see appendix 1). And eventually due to these results, the main goal is to expand the average life expectancy of these animals in the Netherlands.

This paper evaluates the results of the rabbits submitted to this project from November 2010 to February 2011.

Regarding causes of death in these rabbits (hypotheses 1-5) and the relationship between animal welfare issues and disease (hypotheses 6-11) the following hypotheses were formulated:

1 *The most common cause of death of pet rabbits is VHD.*

First of all, there was a closer look at the most common cause of death. In the results of the retrospective research of L.S. Banga [1], VHD was number one cause of death.

2 *Most pet rabbits died of an infectious agent.*

The comparisons that were made started with a first dividing, cause of death: was it infectious, non-infectious or an unknown etiology?

3 *Most rabbits in the ‘group of infectious cause of death’ died by an infection of VHD.*

Followed by this first dividing, there was a closer look at what the cause of death was in the group of infectious cause of death.

4 *Dental problems were the main cause of death in the group of ‘non-infectious causes of death’.* Dental problems are a well-known problem in pet rabbits

[4,7,11,12,14,21,22,29,30,34], therefor the expectation that pet rabbits which didn’t die of an infection, properly died of a dental problem.

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5 *The intestinal organs are the most damaged and/or abnormal in the group of 'unknown etiology' cause of death.* The intestinal tract of the rabbit is a sensitive organ [4,11,12,19,30,32]; therefore the expectation is that these organs are most easily damaged.

6 *Vaccination against VHD and myxomatosis effectively prevents occurrence of these diseases:*

There has been made a comparison between the cause of death is VHD or myxomatosis in relation with vaccination (vaccination against VHD and myxomatosis) statistics. How many rabbits that were vaccinated died of VHD or myxomatosis, and how many that weren't vaccinated died of VHD and myxomatosis?

7 *Outdoor housing of rabbits is related to higher prevalence to infectious disease:*

Some suggest that an animal that is housed inside is less exposed to infectious agents. [7,29] Therefore fewer animals that are housed inside should die of an infectious agent. This is the reason that there has been made a comparison between these two groups. How many rabbits died of an infectious agent and were housed inside, outside or both?

The hypotheses 8 and 9 were inspired by the following information:

Pathogens cause a great deal of responses in vitro, which include a set of immunological responses:

B-cells synthesising Ab (antibodies) which bind to Ag (antigens) therefore facilitating Ag ingestion by macrophages and granulocytes or destruction by complement.

T-cells which destroy cells with Ag on their cell membranes. Natural killer-cells which eliminate cells that lack normal antigens, such as those infected with viruses or tumour cells.

Memory cells which stimulate humoral and cell mediated responses.

Glucocorticoids have many important roles in body regulating functions and in adaptive brain processes. Cortisol is produced in emergency situations where coping is a problem. Results show that high levels of cortisol decrease the immune system:

Decrease of interleukin-1 synthesis by macrophages and of interleukin-2 by T-helper cells causing a lower B-lymphocyte and cytotoxic T-cell activity.

Decrease of interleukin- β that regulates T-helper cell-1 and -2 balance and action.

These effects of cortisol can eventually result in generally worse effects of respiratory pathogens [2,12].

8 *More rabbits died of respiratory diseases which were housed individually than rabbits which were housed in pairs.*

In the study of Chu 2003, there is a comparison between individually housed rabbits and rabbits housed in pairs. Abnormal behaviour is known as an indication of stress [2,6,7,8,9,16,27,33]. The rabbits which were stationed individually, showed a 2 times as high ratio of abnormal behaviour than the paired stationed rabbits. As described above, there seems to be a relationship between an increased level of cortisol and a worse effect of respiratory pathogens [2,26].

9 *More rabbits died of an infection housed in a cage smaller than a floor space of 0,79m² and a height of 0,59m:*

In the research of Dixon et al 2010, there was concern about the pet rabbit hutch sizes. They suggest that the floor area should be at least 0,79m² and the height should be at least 0,59m

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high, for the rabbit to show its natural behaviour [8,9]. In smaller dens, the rabbits show frustrated and stereotype behaviour, which are indicators of stress and have effect on welfare. Because stress reduces the immune system [2,19], an infection with a pathologic agent could be lethal.

10 Rabbits which died of respiratory problems were more frequently housed on wood shaving than other bedding material:

In rats, it is well known that respiratory problems can be induced by wood shavings [30]. Rabbits seem to be housed frequently on wood shavings. Therefore a comparison between wood shaving as bedding with the rabbits which had respiratory diseases as cause of death and rabbits that died of another cause of death

11 Clinical diagnoses made by the treating veterinarian completely corresponded with pathological diagnoses.

Not only the owner provides zootechnical information, the veterinarian also provides information about the anamnesis and a clinical diagnosis. This diagnosis of the veterinarian is based on results of clinical examination which have a limitation. Owners are more reserved when further diagnostics methods for their pet rabbit are proposed, like an X-ray or blood analysis. Certain diseases show limited symptoms or only on the end stage of the disease. And the rabbit is a prey animal, which show limited symptoms when they are ill [13,20,28,30]. All this together makes it harder for the veterinarian to make a good clinical diagnosis. The pathological examination is more expanded, so trauma, malfunctions, inflammations and infections are easier to be found. By comparing the results of the necropsy report (appendix 2) of cause of death with the clinical diagnosis, can provide an insight whether the veterinarian had a good indication or not. It's thinkable that not only welfare problems promote premature death, but a diagnosis made too late or not at all because of limitations has its influence as well.

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Material and Methods:

- Assisting in the *Welfare research small mammals* (rabbits, guinea pigs, rats and ferrets) of the, Department Pathobiology, December 2010 – March 2012: Sampling/transferring the data from November 2010 to February 2011 in the database.
- Comparing the cause of death with the welfare results.
- Literature research
- Comparing the results of rabbits in the *Welfare research small mammals* with the retrospective research of L.S. Banga.

Assisting in the Welfare research small mammals (rabbits, guinea pigs, rats and ferrets) of the, Department Pathobiology, December 2010 – March 2012: sampling and data in database from November 2010 to February 2011.

First the owner of the deceased rabbit completed an extensive anamnesis form (appendix #) regarding zootechnical aspects and medical history. The carcass of the rabbits needed to be cooled (4°C) immediately and within 24 hours post mortem offered for necropsy at the Department Pathobiology of the Veterinary Medicine Faculty at the University of Utrecht. For the necropsies of the carcass there is a protocol especially for the welfare research. First there is an evaluation of the macroscopic aspect of the carcass and organs, samples of organs are collected in 10% neutral buffered Formalin and for freezing (-20°C). Second there is a cytological examination of the liver, spleen, lungs and hindgut and it's contending. If there is an indication in the macroscopic evaluation, like a purulent inflammation of an organ, a bacterial culture was done. Formalin fixed material was routinely paraffin embedded subsequently, 3 micron slides were standard Haematoxylin and Eosin (HE) stained and evaluated by dr. IJzer or dr. Kik, certified veterinary pathologists.

The results of rabbits in the welfare research small mammal, necropsy results compared with the welfare results

All gathered data was inserted in a special database designed by Baijens in 2008 (appendix 3.1, 3.2, 3.3). In this database all data from the extensive zootechnical information that the owner had provided (appendix 1.2) was inserted. When data were missing, owners and/or the treating veterinarian were contacted. Initially, vaccination status of the rabbits missed. By contacting all owners and/or the veterinarian, the omitted vaccination status was gathered. Determined was if the cause of death was infectious, non-infectious or an unknown etiology. The database of Bijens made it possible to export all records to excel. This way we had a clear view of all the inserted data and comparisons were more easily made.

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Hypotheses:

1 *The most common cause of death of pet rabbits is VHD.*

In excel 2010 the data of 'cause of death' was sorted to a high-low rating. The results were used for figure 1.

2 *Most pet rabbits died of an infectious agent.*

In excel 2010, the number of infectious, non-infectious and unknown cause of death were compared. The results were used for figure 2.

Followed by this first dividing, there was a closer look at what the cause of death was. In the group of infectious, 3 *in the 'group of infectious cause of death', most rabbits died by an infection of VHD.*

In excel 2010; the data of 'cause of death' in the group 'infectious' was sorted in a high-low rating. The results were used for figure 3.

4 *dental problems was the main cause of death in the group of 'non-infectious causes of death'* In excel 2010; the data of cause of death in the group 'non-infectious' was sorted in a high-low rating. The results were used for figure 4.

5 *the intestinal organs are the most damaged and/or abnormal in the group of 'unknown etiology' cause of death* In excel 2010; the data of most damaged organs were sorted and put into a high-low rating. The results were used for figure 5.

6 *Vaccination against VHD and myxomatosis effectively prevents occurrence of these diseases:*

In excel 2010; the data of the number of rabbits that were vaccinated was compared with the number of rabbits that weren't vaccinated. In these two groups there was a closer look if any of the rabbits had died of either myxomatosis or VHD. The results were used for figure 6 and 7.

7 *Outdoor housing of rabbits is related to higher prevalence to infectious disease:*

In excel 2010; the data of rabbits that were housed outside was compared with the data of rabbits that died of an infectious agent. The same was done with the data of the rabbits that were housed inside. The results were used for figure 8.

8 *More rabbits died of respiratory diseases which were housed individually than rabbits which were housed in pairs*

In excel 2010; the data of rabbits that died of an respiratory diseases was compared with the data of rabbits that were housed individually. The results were used for figure 9

9 *more rabbits died of an infection housed in a cage smaller than a floor space of 0,79m²:*

In excel 2010; the data of the floor space of all rabbits was sorted, and data of rabbits, no matter how many rabbits, that were housed in a cage with a floor space that was smaller than 0,79m² was compared with the data of rabbits that died of an infectious cause of death. The results were used for figure 10

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10 rabbits which died of respiratory problems were more frequently housed on wood shaving than other bedding material:

In excel 2010; the data of rabbits that died of a respiratory disease was compared with the data of the bedding these rabbits were housed on. The results were used for figure 11.

11 Clinical diagnoses made by the threatening veterinarian completely corresponded with pathological diagnoses.

In excel 2010; the data of all rabbits the 'cause of death' was compared with the data of clinical diagnosis of these rabbits made by the veterinarian. The results were used for figure 12 and 13.

Literature research:

The literature was collected from medical books and online databases; Science direct, PubMed, and Omega. The Goal of the literature research was to determine the most common causes of premature death in pet rabbits and to obtain reliable information about welfare. What, according to scientifically based results, is welfare problems that participate in the cause of death of premature died pet rabbits? We compare these literature results with the results of the extensive zootechnical information that is gathered in the present project.

Comparing the results of rabbits in the 'Welfare research small mammals' with the retrospective research of Drs. L.S. Banga:

Drs. L.S. Banga, performed a research into all necropsy reports of the pet rabbits that were submitted for post mortal examination to the Department Pathobiology of the Veterinary Medicine Faculty at the University of Utrecht from the period '93-'09. Drs. L.S. Banga evaluated only the necropsy reports of pet rabbits between 6 and 36 months old and were kept as pet, so rabbits who were kept for breeding or laboratory research were excluded. In total there were 143 out of 1841 rabbit's useable. The results of the retrospective research will be compared with the results of the welfare research small mammals (pet rabbits) [1]

If there is an indication, the samples that are frozen can always be used for further examination. That this is useful proved itself. 9 rabbits which died of (broncho)pneumonia were suspicious of *Pasteurella multocida* [18,19,20,26,30], therefore a bacterial culture was set with the frozen lungs.

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Results

Total rabbits submitted n=38.

23 female, 4 neutered, 20 intact

14 male, 4 neutered, 10 intact.

Average age of death: 18.9 Months, varies from 4 to 34 months.

1 The most common cause of death of pet rabbits is VHD:

In total there were 38 rabbits, which fitted in the protocol of the welfare research. The results of the necropsy reports gave an insight of the cause of death. As is shown in figure 1 below, the most common cause of death was (broncho)pneumonia (n=9) of which we assumed that it was an infection of bacterial origin. Followed by an encephalitozoon cuniculi infection (n=8).

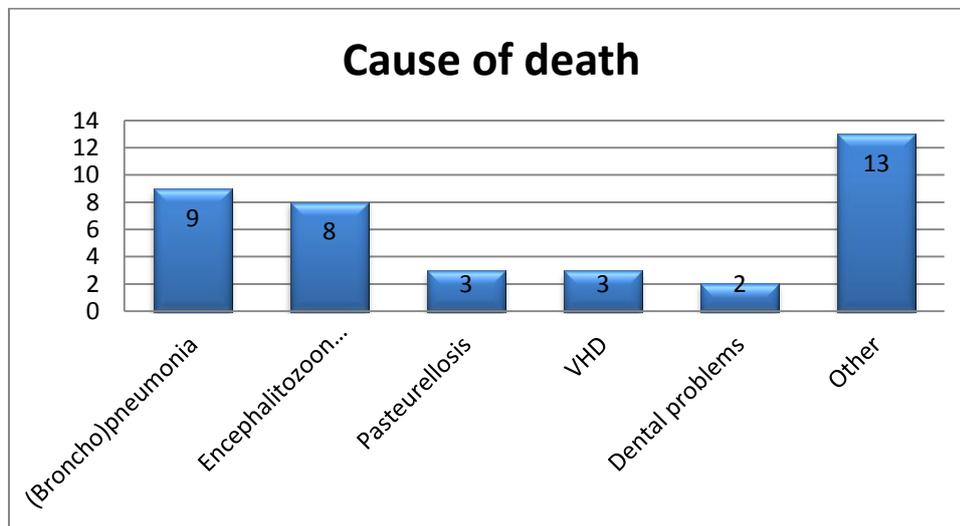


Figure 1: distribution in cause of death of all rabbits of the welfare project.

2 Most pet rabbits died of an infectious agent

Second to the cause of death, there was a determination whether the cause of death was infectious, non-infectious or an unknown ethology. The results of this division:

Infectious: 71.1% (n=27), Non-infectious: 21.0% (n=8), Unknown ethology: 7.9% (n=3)

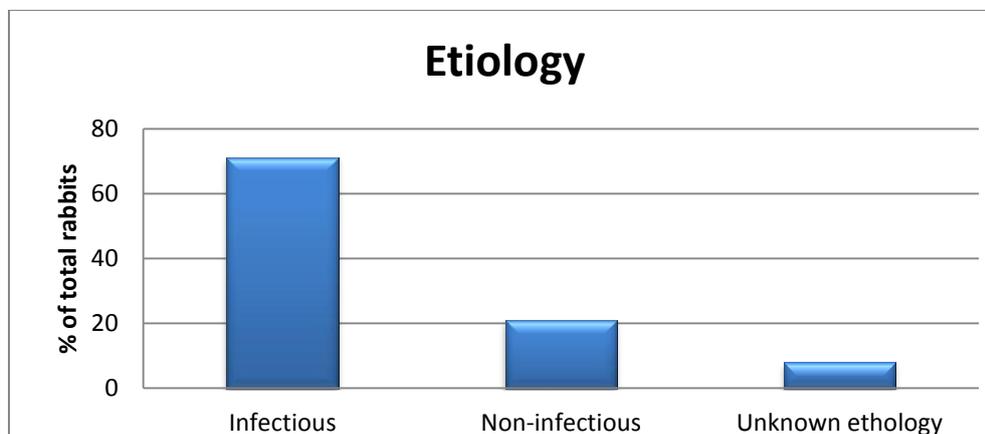


Figure 2: distribution of cause of death by infection, non-infectious or unknown ethology.

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3 Most rabbits in the 'group of infectious cause of death' died by an infection of VHD:

In the group of infectious death, (broncho)pneumonia was the number one cause of death, 33,3% (n=9). Followed by encephalitozoon cuniculi, 29,6% (n=8), and a shared third place for Pasteurellosis, 11,1% (n=3) and VHD (viral haemorrhagic disease), 11,1% (n=3). The results are shown in figure 3.

Together, (broncho)pneumonia and encephalitozoon cuniculi were responsible of 63.0% of death caused by an infectious agent and 44,7% of death cause of all rabbits that participated in the welfare research from November 2010- February 2011.

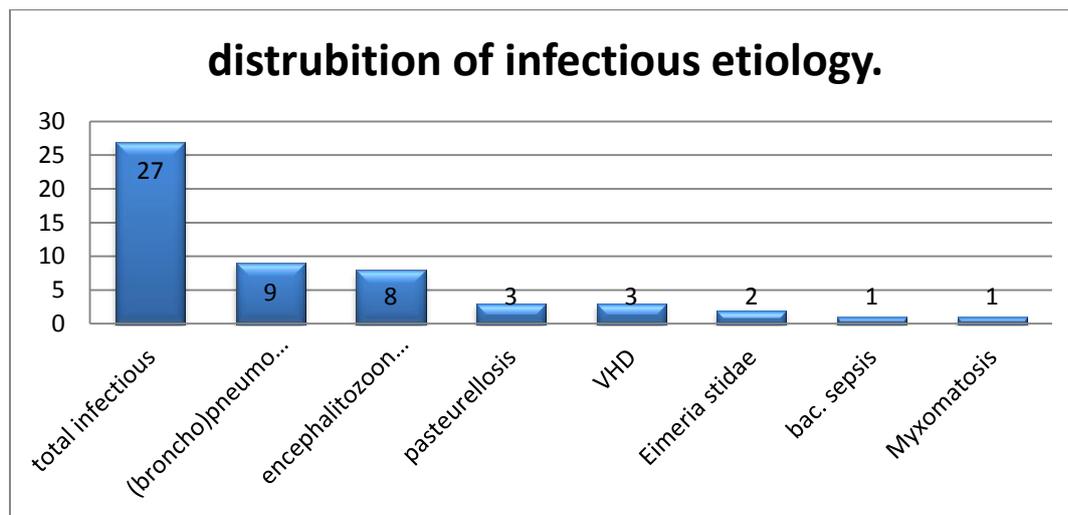


Figure 3: distribution of cause of death by an infectious agent.

4 Dental problems were the main cause of death in the group of 'non-infectious causes of death':

In total, 8 rabbits died of a non-infectious cause of death. 25% (n=2) rabbits died of severe dental problems. The other rabbits died of a number of reasons, as shown in figure 4.

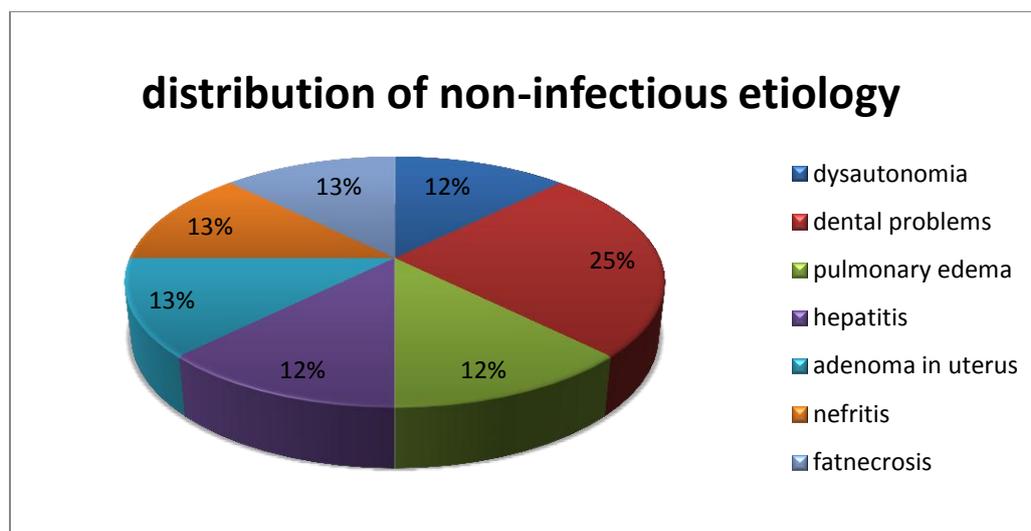


Figure 4: distribution of non-infectious etiology.

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5 The intestinal organs are the most damaged and/or abnormal in the group of 'unknown etiology' cause of death: 3 rabbits were classified under the 'unknown etiology cause of death'. The organs that were most damaged with these three rabbits were the lungs in 2 rabbits and the duodenum of one rabbit.

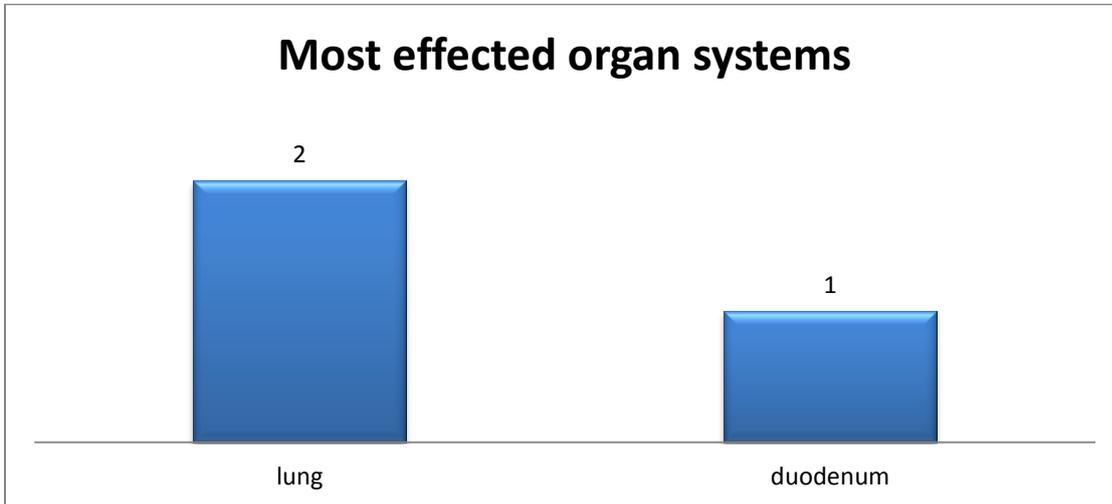


Figure 5 distribution of which organs were most effected in the unknown etiology group.

6 Vaccination against VHD and myxomatosis effectively prevents occurrence of these diseases:

7,9% (n=3) of all rabbits died of an infection with VHD. 2,6% (n=1) of all rabbits died of an infection of myxomatosis. Result show that 63.2% (n=24) was vaccinated, 26.3% (n=10) was not vaccinated and 11% (n=4) is unknown. In the group that was vaccinated, none of them died of VHD or myxomatosis. This is the same in the group that is unknown. In the group that wasn't vaccinated (n=10), 30% (n=3) of them died of VHD and 10% (n=1) died of myxomatosis.

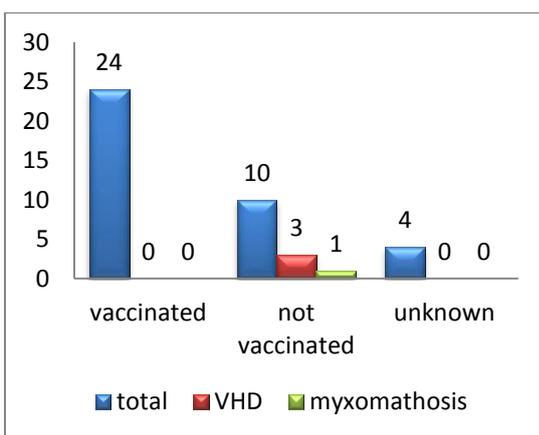


Fig. 6 distribution of death cause by myxomatosis or VHD in the groups vaccinated, not vaccinated or unknown.

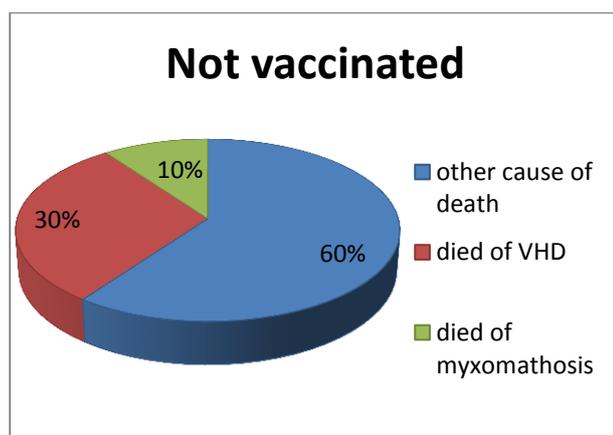


Fig.7 percentage of not vaccinated rabbits that died of myxomatosis or VHD.

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7 Outdoor housing of rabbits is related to higher prevalence to infectious disease:

In total, there were 19 rabbits housed inside, 14 of them died of an infectious cause of death, this is 74%. There were 14 rabbits housed outside, 11 of them died of an infectious cause of death, 73%. 4 Rabbits were held both inside as outside, 2 of them died of an infection, 50%. As is shown in figure 8.

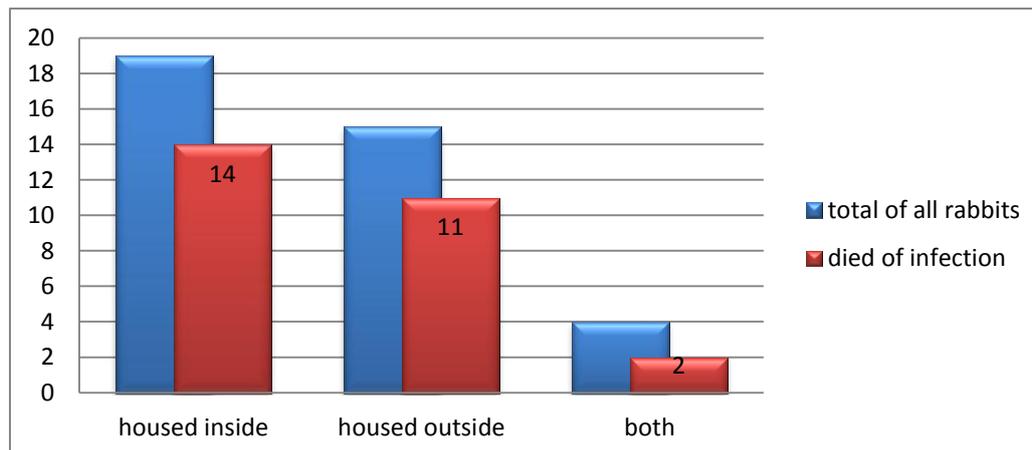


Figure 8: infectious cause of death in the groups; housed inside, outside or both

8: More rabbits died of respiratory diseases which were housed individually than rabbits which were housed in pairs:

43% (n=6) of the rabbits that was housed individually died of a respiratory disease, in paired housed rabbits, this was 10% (n=1). As is shown in figure 9.

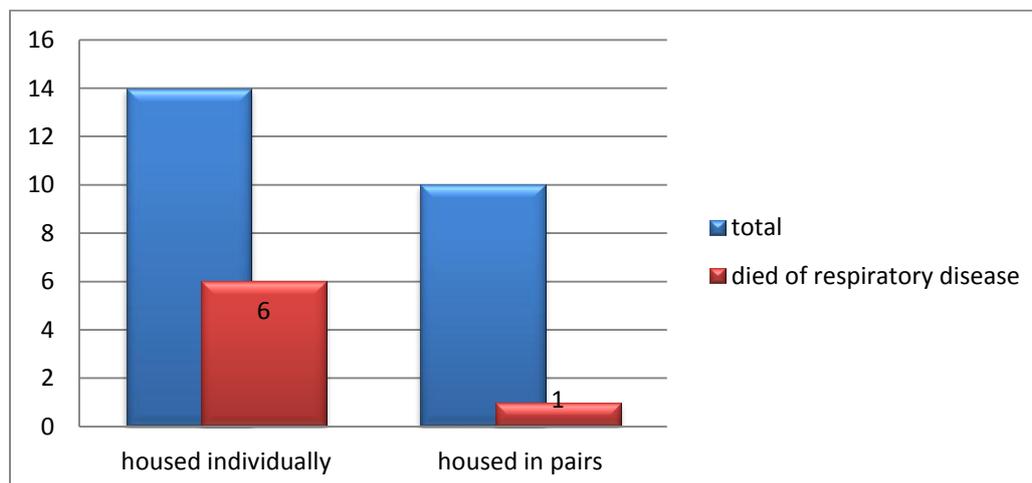


Figure 9: the amount of respiratory diseases in individually housed rabbits versus paired housed rabbits.

9 More rabbits died of an infection housed in a cage smaller than a floor space of 0,79m² and or a height of 0,59m.

18 rabbits out of 38 were housed in a cage which was smaller than 0,79m² or a height of 0,59m. 10 of the 18 rabbits that was housed in smaller cages died of an infectious agent. 20 rabbits were housed in a larger cage than 0,79m² and height of 0,59m and 17 died of an infectious agent. As is shown in figure 10.

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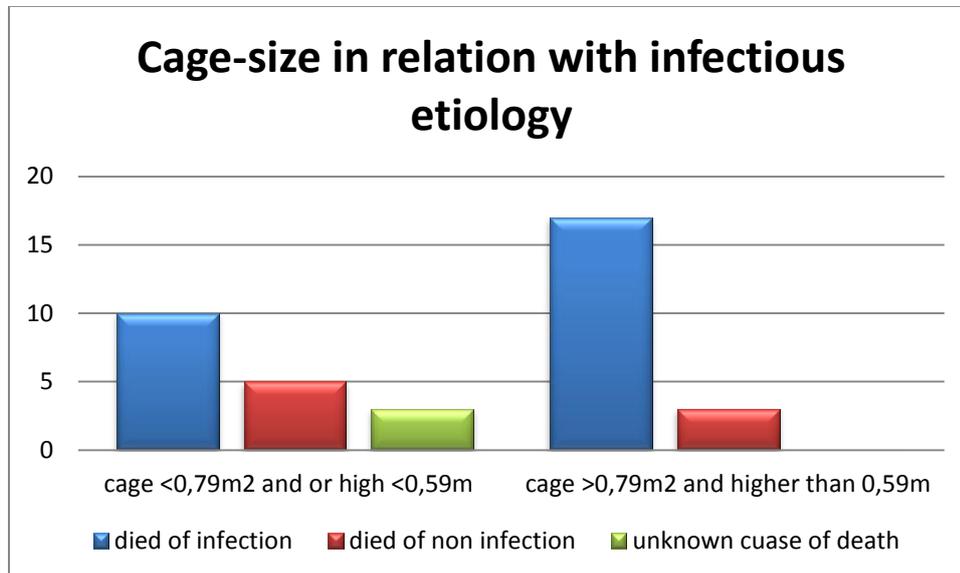


Figure 10: relation between infectious cause of death and the size of the cage.

10 rabbits which died of respiratory problems were more frequently housed on wood shaving than other bedding material:

As said before, (Broncho)pneumonia is the number one cause of death. There was made a comparison between wood shaving as bedding with the rabbits which had respiratory problems and rabbits that died of another cause of death. 70% (n=7) of the rabbits that died of respiratory diseases was bedded on wood shavings. The rabbits that died of other reasons, 43% (n=12) was bedded on wood shavings. In overall, 50% (n=19) was bedded on wood shavings. As is shown in figure 11.

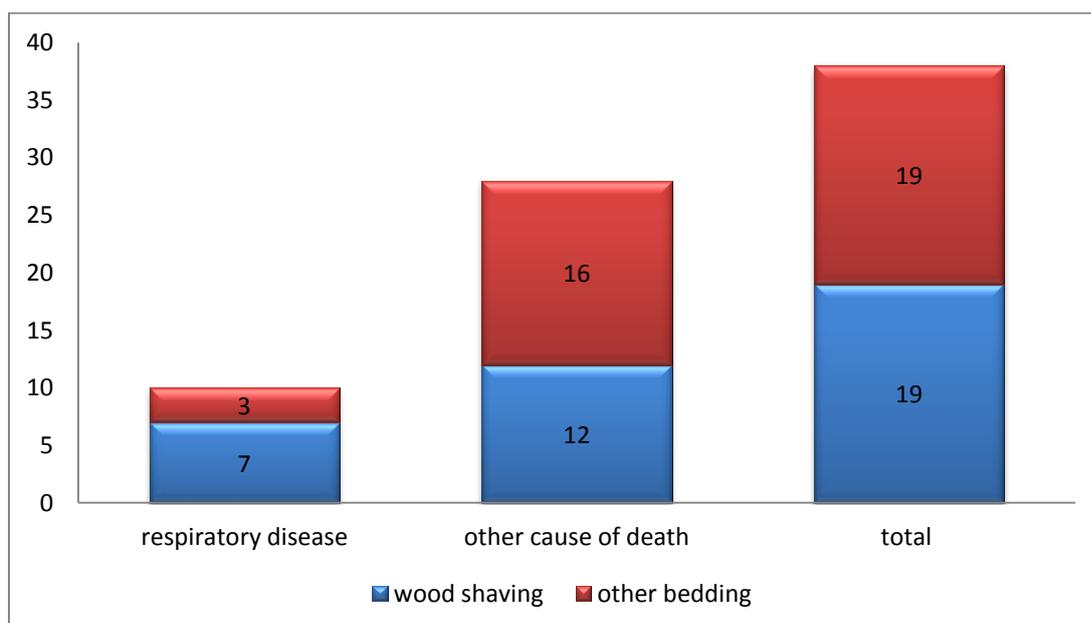


Figure 11: distribution of bedding, wood shaving or other in the groups; respiratory disease cause of death, Other cause of death and the total amount of rabbits.

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11 Clinical diagnoses made by the threatening veterinarian completely corresponded with pathological diagnoses.

In total, 16 out of 38 veterinarians had a clinical diagnosis, 22 out of 38 veterinarians did not have a clinical diagnosis. Out of 38 veterinarians, 8% (n=3) had the correct clinical diagnosis. 34% (n=13) had the incorrect clinical diagnosis and 58% (n=22) did not have a clinical diagnosis at all. Results are shown in figure #. Of the veterinarians that did have a diagnosis, only 19% (n=3) had the correct clinical diagnosis, and 81% (n=13) had the incorrect clinical diagnosis, as is shown in figures 12 and 13 below.

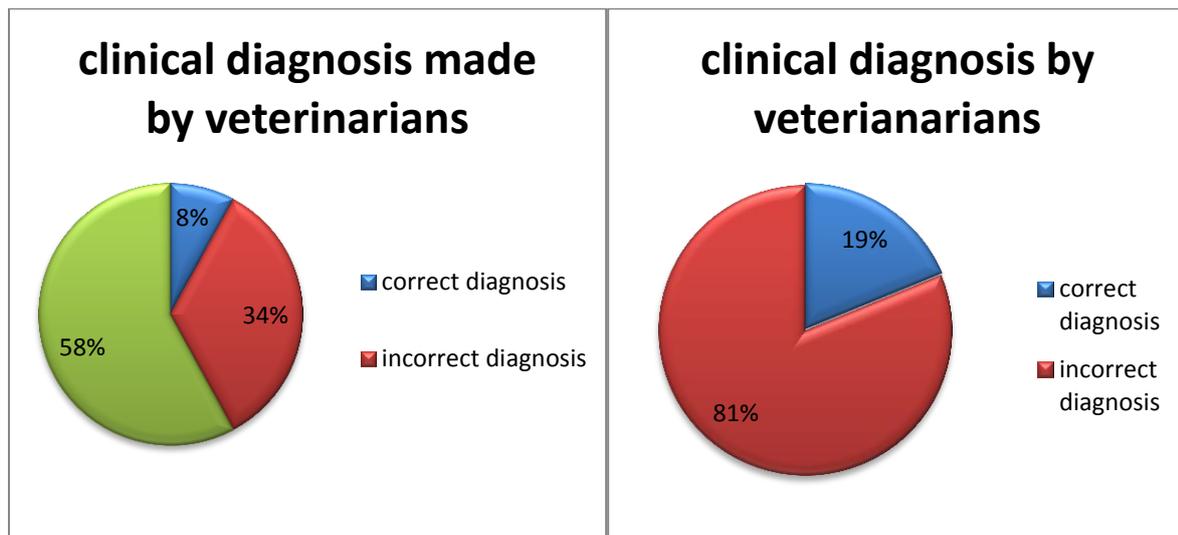


Fig. 12: percentage of all the clinical diagnosis which were Correct, incorrect or unknown (green).

Fig.13: percentage the correct and incorrect clinical made by the veterinarians.

Comparison the results of the welfare research small animals with the retrospective research of drs. L. Banga:

In total there were 143 out of 1841 rabbits who fitted in the protocol for the welfare project. The results of this research was that 67,1% of the pet rabbits died of an infectious agent, 18,2 % died of non-infectious agents and 14,7% died of an unknown etiology. These results were compared with the results of the welfare project from November 2010-february 2011.

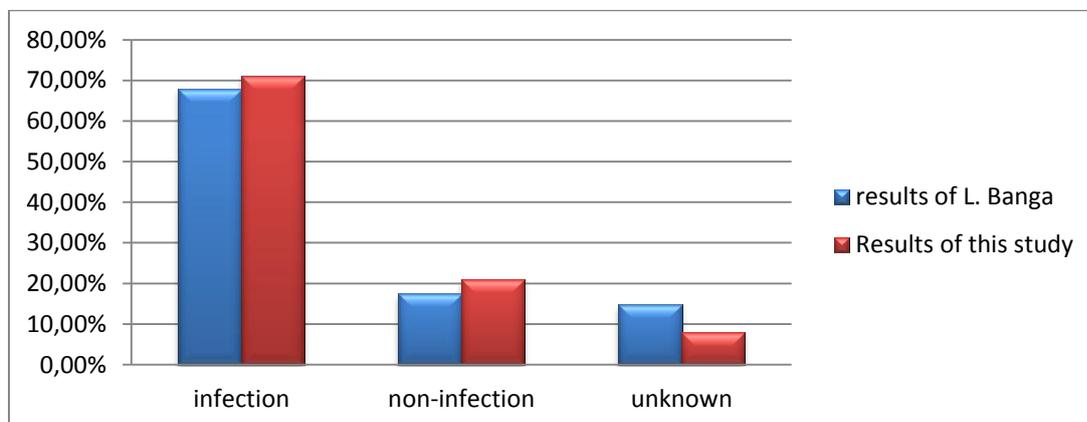


Figure 14: overview of infected versus non infected and unknown cause of death in the research of drs. L. Banga compared with the results of the welfare project in the period of November 2010-february 2011

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In the group of infectious agents, 35,5% had Viral haemorrhagic disease (VHD), 14,0% had *Pasteurella multocida* and 7,0% unspecified bacterial sepsis. As is shown in figure 15.

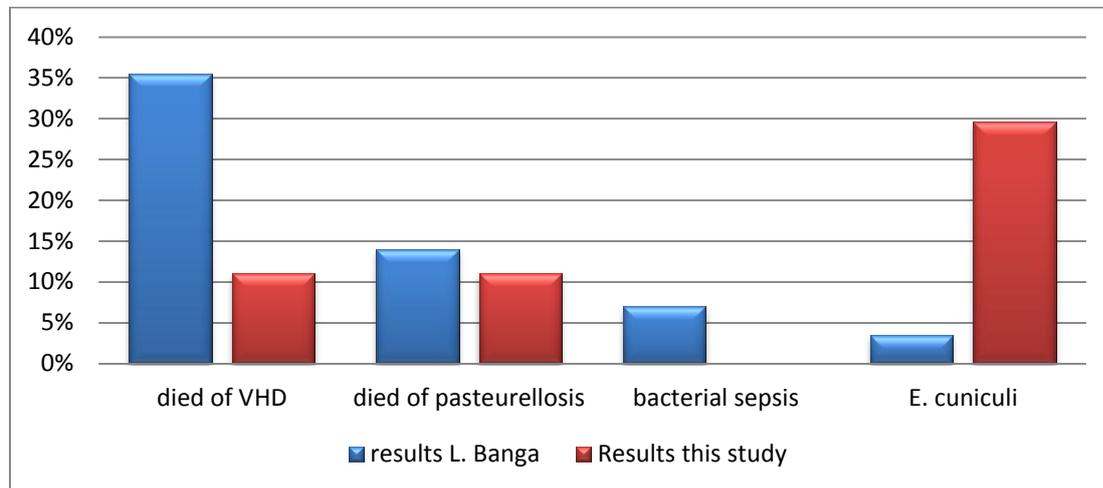


Figure 15: overview of cause of death in infectious causes in the research of drs. L. Banga compared with the results of the welfare project in the period of November 2010-february 2011.

Test results of the bacterial culture of the lung from rabbits that died of a bronchopneumonia. Results of the bacterial culture test were that none of them had a positive test result with *Pasteurella multocida*. The tests showed multicultural bacterial colonies.

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Discussion:

Limitation of this study:

The number of rabbits that is used (n=38) is too small to make waterproof conclusions. If one rabbits dies of a certain disease, 2.6% of the total rabbits died of this disease. To be representable for all rabbits in the Netherlands, the number of rabbits should be much larger. But when the results of drs. L. Banga are compared with the results in this research, the percentage of rabbits that died of an infectious agent is almost equal (67,1% versus 71,1%). This is also the case with the number of rabbits that died of a non-infectious cause of death. In the study of drs. L. Banga were far more rabbits used that in this study (143 versus 38) and due to the equal results it sustains the results in this study.

Further limitation to this study is the way rabbits are gathered. The carcass of the rabbits needed to be cooled (4°C) immediately and within 24 hours post mortem offered for necropsy at the Department Pathobiology of the Veterinary Medicine Faculty at the University of Utrecht by the owners. For most owners this is a big effort and this enlarges when an owner lives further away. Therefore most rabbits were offered from around Utrecht, as result a regional presentation.

Maybe veterinarians are less motivated to offer a rabbit for necropsy when the diagnosis is already made. It is thinkable that more common causes of death or diseases which represent themselves with typical symptoms were less seen in this study because of that.

The next limitation is the extensive zootechnical information provided by the owner. Not all information about the zootechnical is always filled in by the owner. Sometimes the veterinarian or the assistant filled in the paper work, with guesses of the required information.

The previous limitation is also the case on the information gathered about the clinical diagnosis that is filled in by the veterinarian. In some cases the assistant or even the owner has filled this in. 58% of the clinical diagnosis weren't filled in at all. The question that rises is, had the veterinarian no clinical diagnosis because he or she doesn't know it, or is the clinical diagnosis missing due to other reasons, time pressure, forgotten or overseen.

Another limitation is the accuracy of the owner when they filled in the extensive zootechnical information. Cages weren't always measured, the exact amount of food and what kind of food wasn't always filled in completely. In some cases the paper work wasn't filled in at all. Contacting the owner afterward led to the information, but due to the delay valuable information could be lost.

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1 *The most common cause of death of pet rabbits is VHD.*

Of the 38 rabbits, 9 died of (broncho)pneumonia and 3 rabbits died of VHD. Therefore the hypothesis is false. One reason that the number of rabbits that died of VHD is lower than expected, can be related with percentage of rabbits that were vaccinated against VHD (63.2%) and none of the vaccinated rabbits died of VHD. The rabbits that weren't vaccinated against VHD (36,8%). Of the rabbits that were not vaccinated against VHD, 30% died of VHD. Based on these results, there is indication that vaccinating rabbits against VHD reduces the number of rabbits that die of VHD, see also hypothesis 6.

The hypothesis is false in this study; VHD was not the most common cause of death.

2 *Most pet rabbits died of an infectious agent.*

71.1% died of an infectious agent. Therefore the hypothesis is true in this study.

3 *in the 'group of infectious cause of death', most rabbits died by an infection of VHD.*

(Broncho)pneumonia was the number one disease in the group of infectious agents with 33,3%. VHD as cause of death was 11,1% of all rabbits in this study. Therefore the hypothesis is in this study is false.

4 *dental problems was the main cause of death in the group of 'non-infectious causes of death'. 8 rabbits died of a non-infectious cause of death. 2 out of 8 rabbits died due to dental problems in this study. Therefore based on these numbers the hypothesis is true, but this is a very low number, and not significant. Further research is necessary. A reason that there were so little rabbits with dental problems is that most veterinarians diagnosed this themselves. So it is imaginable that the veterinarians did not send these rabbits in for necropsy. And therefore more rabbits actually died of dental problems than this study shows.*

5 *the intestinal organs are the most damaged and/or abnormal in the group of 'unknown etiology' cause of death.* There were 3 rabbits in this group. 2 of them had severe long damage and one of them had duodenum damage. The hypothesis is false based on these numbers. But again, the number of rabbits that are used is low and not significant.

6 *Vaccination against VHD and myxomatosis effectively prevents occurrence of these diseases:* 24 rabbits were vaccinated, none of these rabbits died of myxomatosis or VHD. 14 rabbits were not vaccinated, and 4 of them died of myxomatosis or VHD. Based on the results, the hypothesis is true. More research is required, but it seems to be the case that vaccination against VHD and myxomatosis decreases the number of rabbits that die because of these two diseases.

7 *Outdoor housing of rabbits is related to higher prevalence to infectious disease:*

74% of the rabbits that was housed inside died of an infectious agent. 75% of rabbits that were housed outside died of an infectious agent. There seems to be no relation between housing outside and a higher prevalence to infectious diseases.

8 *More rabbits died of respiratory diseases which were housed individually than rabbits which were housed in pairs.* 14 rabbits were housed individually, 6 (43%) of them died of a respiratory disease. 10 rabbits were housed in pairs, 1 (10%) died of a respiratory disease. The hypothesis based on the results is true, and it is a good indication for further research.

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9 More rabbits died of an infection housed in a cage smaller than a floor space of 0,79m² and or a height lower than 0,59m: 18 rabbits were caged in a cage that was smaller than 0,79m² or lower than 0,59m. 10 of those rabbits died of an infection. 20 rabbits were caged in a larger and higher cage. 17 died of an infection. Therefore the hypothesis in this study is false. There should be more comparisons with amount of space and health problems to find the minimal floor space for a rabbit to live healthier.

10 rabbits which died of respiratory problems were more frequently housed on wood shaving than other bedding material: 70% of the rabbits that died of respiratory diseases were housed on wood shavings. 43% of the rabbits that died of other than respiratory diseases were housed on wood shavings. In total of all rabbits, 50% was housed on wood shavings. Therefore there seems to be no relation between wood shavings as bedding and respiratory problems, and the hypothesis is false.

11 Clinical diagnoses made by the treating veterinarian completely corresponded with pathological diagnoses. There were 16 cases who had a clinical diagnosis out of 38. Of these 16 diagnoses there were 3 in correlation with the pathological diagnosis. Therefore the hypothesis in this study is false. But there is a limited number of veterinarians who filled in the paperwork, and no certainty if the veterinarian filled it in or the assistant. But there seems to be evidence that more research is required for a good diagnosis. This statement needs more research, but it is an interesting result.

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Recommendations for pet rabbit owners

Go to the veterinarian to vaccinate your rabbit against VHD and myxomatosis

In our study, it shows that none of the vaccinated rabbits died of these diseases. The retrospective research of drs. L. Banga showed that VHD was number one cause of death from 1993-2009. Therefore it looks like vaccination helps against these lethal diseases. Studies show that there are more ways to vaccinate your rabbit against myxomatosis and VHD, therefore contact your veterinarian for what options there are, and what is best for your rabbit in particular [7,20,28,30,32]

Housing pet rabbits in pairs.

In our study respiratory diseases were number one cause of death. There seems to be evidence that housing rabbits in pairs, reduces stress and therefore are less vulnerable for respiratory diseases. A rabbit is a social animal, which needs a companion to communicate with, to find support in stressful situations, to nurse with each other, to form a union and are together twenty-four-seven. Humans, how good their intentions are, are not rabbits, so the communication will not be the same and will not be in interaction twenty-four-seven with the rabbit [4,6,20,23,28,29,31,33]

Inside or outside housing had no influence on the prevalence of infectious diseases.

There was no evidence that housing rabbits outside leads to a higher risk that the rabbit dies of an infectious agent. So housing rabbits both inside as outside is no problem if it comes to infectious diseases, studies even show that rabbits have less stress when they are outside [7].

Visit the veterinarian when you bought your pet rabbit:

When you get a new rabbit, at the local pet shop or from friends, it is necessary to let your pet rabbit be examined for health problems. Young rabbits are already vulnerable for diseases and when they get a new home it is a stressful period for them and stress reduces the immune system [2,12,20] which makes them even more vulnerable. The veterinarian can examine the young rabbit, and when needed, treat it as well. And most important, there is a lot of information about 'how to keep your rabbit', but at the veterinarian you get the most reliable, based on scientific results, information for your rabbit. How to keep them happy, what kind of food they should get and how much. Information about vaccination, what signs of illnesses there are, when to call the veterinarian, how to cut the nails, how much your rabbit should weight and more.

When your pet rabbit is ill, don't wait and give the veterinarian the opportunity to expand its examination by further research if it is necessary.

First of all, when you see that your rabbit is ill, it is a serious case. Rabbits usually don't show their illness, so when they do, they are really sick. Even when they start to eat less, you should go to the veterinarian immediate. It is not as common with dogs and cats to do further research as X-rays or blood analysis, but exactly these examinations can make the difference between life and death for your rabbit [4,6,20,24,25,32,34].

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Recommendations for veterinarians

Create a young rabbit consultation:

With dogs and cats, it's common to have a first free or with little cost consultation about their young animal. Why not with rabbits? After all it is number three most kept animal in the Netherlands and UK as well. These way owners get the most reliable information about how to keep their rabbits healthy and happy. And in the future an owner probably consults the veterinarian sooner when there is a problem with the rabbit. If the veterinarian makes the rabbit an important animal to come to the vet, owners will soon follow their example.

Create a young rabbit's (e.g. puppy starter kit) starters pack:

A young rabbit's starters pack with evidence based information about how to keep a rabbit healthy and happy. There can be all kind of healthy and fun materials in the starter's pack here are some ideas:

- Food samples of the brand that you prefer and sell for rabbits.
- A schedule card with how much should my rabbit eat and drink
- A schedule card of how much should my rabbit weigh.
- Information about vaccinations against VHD and myxomatosis, what you already told, and the owner can read it at home.
- Information about preventive healthcare. Like neutering female rabbits.
- A list of what kind of side dishes the rabbit may eat, like fruits and vegetables.
- A list of what things that make a rabbit happy, like companions, enough space and more.
- A small book with information about the rabbit in general.
- What kind of diseases is common with the rabbit, and what are their symptoms.
- When to call a veterinarian immediately.
- A rabbit's toy or snack.

The better an owner gets informed with the right information about rabbits; the more rabbits will stay healthy, or in case of illness go to the veterinarian. This way we create a small step that the average age of rabbits in the Netherlands will expand.

If a rabbit is ill and needs further diagnostic work, explain this loud and clear to the owner that it is necessary.

In this study there is evidence that the veterinarians did not have the full picture of what was going on with the rabbit. It is not very common to use further research on rabbits, but it is a growing sector. So talk to the owners about this options and make clear that they are necessary for a good diagnosis and therefore treatment. Emphasise the need of more diagnostic work toward the owner, and create the feeling that their rabbit is worth it.

Become friends with the pet-shop nearby:

What could work to get owners to your young rabbit consultation is to create a coupon, which owners receive when they buy a rabbit at the pet shop. The coupon can be used by owners for a free young rabbit consultation at your practice. Make sure that it expires within a limited time, like a week or so. Otherwise people will use it as soon as the rabbit is ill, and that is not the goal of these coupons. Besides the coupon, many rabbit owners ask information about their rabbit in their local pet-shop. If you work more together, and even educate the employers

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of the pet-shop, more rabbits will get the proper care and, in case of illness, directed to your practice.

Be up to date about which rabbit illness is common at the time.

In the research of L. Banga, VHD was number one disease. E. Cuniculi was not really a problem. In this study, E. Cuniculi is responsible for more than a fifth of all deaths. One clinical diagnosis was correct about this disease. Make sure that as a veterinarian, you keep yourself informed what rabbit illness is common right now [11,32].

Tips in general:

It looks like one of the major problems is that owners do not have enough knowledge about their pet rabbit [4,5,6,10,20,27,29,31,33,34] . Reliable information for owners about their pet rabbit is necessary, but it needs to be easy to find, easy to understand and interesting for the owner to use.

Create short funny but informative videos about how to keep your rabbit healthy:

- How to pick a rabbit up
- What is dangerous in house for a rabbit
- What are signs of illness, like not eating, sticky faeces, slower than normal etc.
- How to give medication to a rabbit
- How to cut the nails of the rabbit
- Why a rabbit should not be overweighed
- Global information about the rabbit, like that is a prey animal, how to prevent stress.
- What normal behaviour is for a rabbit

This is only a rough idea of what kind of videos could be made. These videos can be used by veterinarians on their internet site, or links of them.

Have all the reliable information on a website that cooperates with veterinarians, so that the veterinarian can use this internet site as a link on his own site or recommend it to the owners.

Create a downloadable app for smartphones, which rabbit owners can download free of charge or at a minimum cost, which contains information about rabbits, short and simple but accurate. For example: a food diagram, how much food does my rabbit need, or a diagram about how the rabbit grows, or simple instructions, what to feed and what not and so on.

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Appendixes:

Appendix 1, Anamnesis form provided by the owner and the threatening veterinarian:

Anamnesis form *Welfare research small mammals*

(rabbits, guinea pigs, rats and ferrets)

VPDC, Postbus 80158, 3508TD UTRECHT **Tel. 030- 253 3195** Fax: 030-2534774

Practice:

Veterinarian: Email:

Phone number.:

Owner Rabbit/guinea pig/rat/ferret

Street: Name animal:

Zip code: Date of birth:

City: Gender:

Phone number.: Owner since:

Code owner:

Cremation no/ yes, to

Euthanasia/ **Died** dd:

Onset of illness:

Number of animals in group:

Number of diseased animals in group:

Medication:

Duration of treatment:

Clinical signs Diarrhea Respiratory problems Skin problems

Lameness Depressed growth Sudden death

Loss of appetite/ anorexia Depressed water consumption

Husbandry:

Location: Inside Outside

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Measurements: L: W: H:

Materials: Wood Metal Plastic/Fibreglass
 Glas Other.....

Bedding:.....

Extra ventilation: No Yes

Frequency of cleaning:.....

Detergent type:.....

Frequency of feeding:.....

Food types:

0 Pellets: Brand..... Notes:.....

0 Hay Brand..... Notes:.....

0 Vegetable Type..... Notes:.....

0 Fruit Type..... Notes:.....

0 Snacks Brand..... Notes:.....

0 Other Description.....

Actual intake:.....

Food supplements (incl. vitamin C): No Yes

Water supply: Tap water Bottled water Rain water Other

Type: Bowl Drinking nipple

Frequency water changes:.....

Anamnesis:

Clinical diagnosis:

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Appendix 2: Necropsy protocol *Welfare research small mammals*, VPDC

Welfare project small mammals Rabbit, guinea pig, rat and ferret		
Macroscopy	Microscopy: put in cassettes	Freeze
Identification chip / tattoo		
Body weight		
Skin, nails	Skin left flank	
Bone (right femur)	Right femoral head, decalcify	
Skeletal muscle	Dorsal back muscles, left	
Eyes	OS and OD	
External ears		
Nose, larynx	Conchae	
Mouth, teeth		
Pharynx and oesophagus	Oesophagus	
(para) thyroid	(para) thyroid	
Trachea	Trachea	
Lung	Lung	Lung
Heart	Heart, full circle at 1/3 height	Heart
Large vessels		
		Fat
Intestinal tract	Stomach, duodenum, jejunum, caecum, colon	Colon
Pancreas	Pancreas	Pancreas
Liver and gall bladder	Liver and gall bladder	Liver
Kidney and urinary tract	Kidney, bladder	Kidneys
Spleen, lymph nodes	Spleen, Lnn Mesenteriales	Spleen
Bone marrow (right femur)	Bone marrow right femur, distal and middiafyse	
Adrenal gland	Adrenal gland	Adrenal gland
Brain	Brain	Brain
Pituitary gland	Pituitary gland	
Reproduction organs	Ovaries, uterus, testicles	
Other pathologically changed areas	Other pathologically changed areas	

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Appendix 3-1 Database by Bijens 2008: General form with animal records.

The screenshot shows a FileMaker Pro window titled 'Welzijnsonderzoek_Fretten'. The interface includes a menu bar (Bestand, Bewerken, Weergave, Invvoegen, Opmaak, Records, Scripts, Venster, Help) and a toolbar with navigation and record management icons. The main content area is titled 'Animal Records' and features a sidebar with 'General', 'Zoos & ID', and 'Notes' sections. The 'General' section contains fields for Case No. (0000070), Species (Ferret), Gender (Male), Castration (No), Date of Birth (15-4-1998), Date of Death (12-1-2000), Type of Death (Non-euthanised), Age at death (20 months), Birth type (Captive), Sire ID, Dam ID, and Animal weight (0 grams). The 'Zoos & ID' section includes a Location dropdown (Department), a table for tracking ownership by zoo (Zoo 1 to Zoo 4) with Date In, Date Out, and Zoo ID columns, and fields for Chipnumber and EEP ID (F001). The 'Notes' section has a large text area for special notes. On the right side, there are buttons for 'General List', 'Anamnesis', 'Necropsy', 'Insert Picture', and 'Export Picture'. The status bar at the bottom shows '100' and 'Bladeren'.

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Appendix 3-2 Database: Animal History.

FileMaker Pro
Bestand Bewerken Weergave Invoegen Opmaak Records Scripts Venster Help

Welzijnsonderzoek_Fretten

Records: 1 / 70 Totaal: (Ongesort.)

Alles tonen Nieuwe record Record verwijderen Zoeken Sorteren

Lay-out: Anamnesis History Weergeven als: Schermafdruck Lay-out bewerken

Anamnesis History

New Delete Find Save as PDF Save as Excel

ID in EEP: F001 DoD: 12-1-2000 Mode of Death: Non-euthanised
Case Number: 0000070 Gender: Male Castrated: No

General

Onset of Illness: []
Number of animals in group: [] [] [] [] []
Number of diseased animals in group: [] [] [] [] [] [] [] [] [] []
Medication: []
Duration of treatment: []

General form
General list
Necropsy

Clinical signs

Loss of appetite \ anorexia Skin problems
 Depressed water consumption Lameness
 Diarrhea Depressed growth
 Respiratory problems Sudden death

Husbandry

Location: Inside Outside Extra ventilation: []
Materials: Wood Metal Plastic / Fibreglass Glass Other

Measurements

Space length: [] cm Space width: [] cm Space height: [] cm
Area: [] cm²

Cleaning

Frequency: []
Detergent type: None

Bedding

Hay Straw Wood shavings Wood cat litter pellets Other
Other description: Other bedding material

Nutrition

Frequency of feeding: []
Food supplements: []

Diet

Food types: Pellets Mix Hay Vegetables Fruit Snacks Other
Pellet brand: [] Notes: []
Mix brand: [] Notes: []
Hay brand: [] Notes: []
Veg type: [] Notes: []
Fruit type: [] Notes: []
Snacks brand: [] Notes: []
Notes: []

Actual intake: []

Water supply

Water supply form: Bowl Drinking
Water supply: Tap water Bottled water Rain water Other
Specify: []
Frequency water changes: []

Anamnesis

7.1.2000: opgehaald uit frettenasiel.
Sinds 8.1.2000 sloom.
10.1.2000: behandeld met gluc. 3,14 mmol/l
12.1.2000: epileptiforme aanvallen.
Doorverwezen naar UKG, daar dood bij aankomst.

Diagnosis

[]

100 Bladeren

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Appendix 3-3 Database: Necropsy form

FileMaker Pro

Bestand Bewerken Weergave Invoegen Opmaak Records Scripts Venster Help

Welzijsonderzoek_Fretten

Records: 1 (70 Totaal: (Ongesort.))

Alles tonen Nieuwe record Record verwijderen Zoeken Sorteren

Lay-out: Necropsy Weergeven als: Schermafdruck

Animal Necropsies

New Delete Find Save as PDF Save as Excel

ID in EEP: F001 DoD: 12-1-2000 Necropsy Date: Necropsy Lab: Necropsy Item: Whole body Gender: Male

General Form
General List
Anamnesis

LABEL	ANOMALY?	DESCRIPTION
General Aspect		
Exterior skin	No data	gb
Eyes/ ears	No data	
Orifices mucous memb	No data	
Mouth	No data	
Trachea	No data	gb
Thyroid	No data	een bijschildklier, rechts vrij fors. Linker
Thorax	No data	
Thymus	No data	
Pericardium	No data	
Lungs	No data	longen zijn rose/rood met voldoende luchthoudend
Heart	No data	gb
Abdomen	No data	
Peritoneum	No data	
Stomach	No data	maag bevat veel dik slijmerige inhoud, geen voedsel
Small Intestine	No data	
Duodenum	No data	
Jejunum	No data	Op driekwart van de jejunumlengte bevond zich een
Ileum	No data	
Large intestine	No data	
Caecum	No data	
Colon	No data	
Anus rectum	No data	
Liver	No data	grote lever, geel/rode tekening lijkt macroscopisch
Pancreas	No data	In de pancreas waren een aantal verdikkingen
Kidneys	No data	Rechter bijnier was niet te vinden, nieren
Adrenals	No data	
Spleen	No data	:vrij groot en donker gekleurd, witte
Urine bladder	No data	
Reproductive organs	No data	gb
Joints bones	No data	
Muscles	No data	redelijk tot mager
Lymph nodes	No data	
Vascular system	No data	gb
Nervous system	No data	
Placenta	No data	
Bone marrow	No data	
Mammary glands	No data	

100 Bladeren