

Chemical and Surgical Castration of Male Dogs:

Behavioral Effects

Research Project Veterinary Medicine

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Abstract

Chemical castration using GnRH agonists has proven to be an effective method of rendering dogs infertile as a reversible alternative to orchiectomy. However, little is known about the behavioral effects of chemical castration. In this study, the effects of surgical and chemical castration on aggression, sexual behavior, play behavior and fear/insecurity in male dogs were assessed. In order to do so, twenty-three dogs were chemically castrated using implants which slowly release the GnRH agonist Deslorelin (Suprelorin®), and 18 dogs were surgically castrated. Their behavior was assessed on the day of, but prior to treatment and 4-5 months after treatment by means of a behavior test and a questionnaire that was filled out by the dog-owners.

No significant changes in fear/insecurity and aggression were seen after treatment using the behavior tests in both groups. However, many owners did observe a decrease in aggressive behavior towards other male dogs (suprelorin implant group: 43.5%, surgical castration group: 47.0%). Moreover, no significant differences could be found between groups concerning the individual results of the behavior test and the results of the perceptive questions concerning aggressive behavior and fear/insecurity. Play behavior increased significantly in the suprelorin group as well as the surgical castration group, both groups showed a similar increase ($p=0.008$ and $p=0.041$ respectively). Additionally, the owners of the dogs in both groups observed a decrease of sexual behavior towards estrus bitches. However, significantly more owners of the surgical castration group noticed this decrease, compared to the owners of the suprelorin implant group ($p=0.018$). A decrease in sexual behavior was not observed during the behavior tests.

Overall, this study shows that surgical and chemical castration in dogs induce similar effects with regard to aggression, fear/insecurity and play behavior. However, according to our study, a greater effect on sexual behavior can be expected following orchiectomy compared to the changes after chemical castration.

Although this study provides some interesting information about the behavioral effects of surgical and chemical castration, more research is needed in order to make a clear statement about the degree in which surgical castration and chemical castration lead to similar results.

Introduction

GnRH agonists and male reproduction

Many dogs are surgically castrated each year. The reason for castration is often an attempt to eliminate unwanted behavior such as roaming, aggression and hypersexuality. However, surgical castration does not always lead to a clear improvement of the dog's behavior. Moreover, some owners hesitate to have their dog castrated, since it is an irreversible procedure. Therefore, there is a need for a safe, effective and less invasive method of rendering male dogs infertile instead of surgical castration. Chemical castration by use of a potent GnRH agonist provides such a method. GnRH agonists act by suppressing the pituitary-gonadal axis (Trigg et al. 2006, 1507-1512; Junaidi et al. 2007, 891-898). However, initially, a rise in plasma LH and FSH and subsequently testosterone is seen after administration of the GnRH analogue (Vickery et al. 1985, 53-60; Junaidi et al. 2003, 317-322). When the agonist is administered continuously, this acute response is followed by a gradual decline of LH and FSH, due to desensitisation of the gonadotrophs for GnRH by internalisation of the GnRH receptors (Junaidi et al. 2007, 891-898; Vickery et al. 1985, 53-60; Junaidi et al. 2003, 317-322; Gobello 2006, 1560-1567; Junaidi et al. 2009, 725-734; Inaba et al. 1996, 671-677). As LH concentrations drop, Leydig cells in the testes secrete significantly less testosterone. Consequently, the plasma testosterone concentration decreases to basal levels within 6 weeks resulting in spermatogenic arrest and decreased testicular and prostatic volume, apparently in a dose dependent manner (Vickery et al. 1985, 53-60; Junaidi et al. 2009, 725-734).

The effects of chronic GnRH analogue treatment are completely reversible. Restoration of gonadal steroid synthesis inducing spermatogenesis that is associated with fertility gradually occurs once treatment has been stopped (Inaba et al. 1996, 671-677). Eventually, full recovery will take place and the dogs will be as fertile as they were prior to treatment (Trigg et al. 2001, 255-261). The onset of recovery is dose-dependent, dogs treated with a high dose of GnRH agonist will not recover as fast as dogs treated with a low dose (Junaidi et al. 2009, 725-734).

A great variety in the effects of GnRH agonists among different species can be observed. GnRH agonist treatment will (after an initial stimulation) suppress testosterone concentrations in male dogs, male ferrets (Schoemaker et al. 2008, 161-167), male mice (Kher and Kalla 1996, 299-306), rams (Lincoln, Fraser, and Abbott 1986, 587-597) monk seals (Atkinson et al. 1998, 178-182), rhesus monkeys (Sundaram et al. 1984, 365-371) boars (Schneider et al. 1998, 69-80; Xue et al. 1994, 1290-1298), rats (Belanger et al. 1980, 1094-1101) and stallions (Johnson, Thompson, and Cartmill 2003, 1300-1307). In some species of animals, including the wallaby, GnRH agonists do not affect the testosterone concentrations (Herbert et al. 2004, 1836-1842) or may even lead to an increase in testosterone concentration. This is seen in bulls (D'Occhio and Aspden 1996, 45-52; Ronayne, Enright, and Roche 1993, 179-189).

In our study, a group of dogs was treated with implants slowly releasing the GnRH agonist deslorelin (Suprelorin®). Deslorelin is a super GnRH agonist, with a potency of about 10-144 times that of native GnRH (Padula 2005, 115-126). Two different formulations are available: a 4.7 mg implant and a 9.4 mg implant. The effect will last for a minimum of six or twelve months in the 4.7

and 9.4 mg implants respectively, with a large variation. Suprelorin® has been registered in the EU for the indication of the induction of temporary infertility in healthy, sexually mature male dogs (European Medicines Agency 2010).

The Suprelorin® implant should be administered subcutaneously, between the lower neck and lumbar area. Moderate swelling may occur following administration of the implant. Also, there may be some local reaction for up to three months after implantation (European Medicines Agency 2010).

Behavioral effects of surgical castration

Castration is often performed in an attempt to modify the dog's behavior in one or multiple ways (Hopkins, Schubert, and Hart 1980, 1108-1110). Multiple studies have shown that surgical castration can indeed be an effective method to change a dog's behavior. Various behavioral patterns can be altered by surgical castration, in this introduction we will focus on the effects on aggression, sexual behavior, play behavior and fear.

Aggression

Aggression is one of the behaviors which is most mentioned as a motivation for castration of a male dog (Maarschalkerweerd et al. 1997, 617-619). Aggression is a complex behavior. Overall, males are generally more aggressive than females (Goddard and Beilharz 1983, 299-315; Wright and Nesselrote 1987, 169-178) and in many species a relationship between testosterone levels and aggression was found (Giammanco et al. 2005, RA136-45). Although many studies have been published which discuss the effects of surgical castration on aggressive behavior, their results are not always consistent. When looking at aggressive behavior, it is important to consider that aggressive behavior can be divided into different types of aggression.

Various interview-based studies have assessed the effects of surgical castration on inter-male aggression. A study by Hopkins et al. showed that out of 8 dogs displaying aggression towards other male dogs, 5 dogs (62.5%) showed a decrease of this behavior after surgical castration (Hopkins, Schubert, and Hart 1980, 1108-1110). This percentage of decrease is quite similar to the decrease found by Maarschalkerweerd et al. In his study, he found that decrease of inter-male aggression in 57% (n=23) of the dogs following castration (Maarschalkerweerd et al. 1997, 617-619). However, an increase of this objectionable behavior was reported in one dog. Beaver (1983) found that surgical castration lead to a decrease of inter-male aggression in approximately 75% of the cases (Beaver 1983, 35-43). Another study shows that inter-male aggression is reduced or eliminated in 50-60 % (n = 42) of dogs following castration (Hart 1979, 461-465).

Another type of aggression which is a common problem in male dogs is competitive (dominance) aggression. This aggression is seen between dogs or between human and dog in order to establish social position. Theoretically, this behavior should not be altered by castration, since it is a learned behavior with little hormonal control (Beaver 1983, 35-43). However, research has shown that in some dogs, castration does lead to a decrease in competitive aggression. This might be a reflection

of a high or total socialization to humans. As a result, the dog will treat humans as they would treat another dog. If this is true, in such cases, competitive aggression might actually be a variation of inter-male aggression and thus be responsive to castration (Beaver 1983, 35-43).

Little research has been done concerning the effects of surgical castration on other types of aggression. Most of the published studies only focus on inter-male aggression. However, in some studies various types of aggression were reviewed.

Maarschalkerweerd divided aggression into aggression outside, aggression inside, aggression towards unfamiliar people, aggression towards familiar people, inter-male aggression, aggression towards bitches and aggression towards other objects. Twenty-three dogs were included in this study. Of these dogs, 52% showed a decrease of aggression outside, 26% showed a decrease of aggression inside, aggression towards unfamiliar people decreased in 22% of the dogs, 9% of the dogs showed a decrease of aggression towards familiar people, 26% showed a decrease of aggressive behavior towards bitches. No decrease of aggression towards other objects was found. Although many dogs showed a decrease of aggression in certain situations, some dogs showed an increase of aggression. One dog showed an increase of aggression outside the house, towards unfamiliar people and towards familiar people (Maarschalkerweerd et al. 1997, 617-619).

Neilson also split up aggression into different categories and found that with regard to aggression towards other canine or human family members, approximately 25% of the dogs will have a 50-90% improvement after surgical castration. In 10-15% of the dogs, a comparable reduction in aggression towards unfamiliar dogs and humans can be expected (Neilson, Eckstein, and Hart 1997, 180-182).

The study of Hopkins et al. primarily focused on inter-male aggression, but they also studied territorial aggression (n=8) and fear-induced aggression (n=4). None of the dogs showed a decrease (or an increase) in territorial aggression or fear-induced aggression following castration (Hopkins, Schubert, and Hart 1980, 1108-1110).

In a study performed by Blackshaw (1991), castration (alone, or in combination with progestins and/or obedience training) proved to be an effective treatment for certain dogs that showed dominance aggression, territorial aggression, predatory aggression, inter-male aggression, sibling rivalry or fear biting (Blackshaw 1991, 351-361).

The information provided above shows us that there is still a lot of controversy concerning the effects of orchietomy on canine aggression.

Sexual behavior

One of the most common mentioned motivations for castration is unwanted sexual behavior. Sexual behavior in male dogs can be divided into several elements: appetitive responses, such as genital investigation and mounting, and consummatory acts such as erection and ejaculation. When looking at the effects of castration, it is useful to separate these responses, since a change in mounting and other precopulatory behavioral elements may reflect a reduction in the male's sexual motivation, while impairment in sexual potency may reflect impairment in erection or ejaculation (Hart 1974, 383-400).

Several studies have been performed during which the appetitive responses following surgical castration have been assessed. Nearly all of these studies found that surgical castration in dogs leads to a reduction of mounting. Several studies found that orchietomy leads to a reduction of mounting in 50 to 70 percent of the treated dogs (Hopkins, Schubert, and Hart 1980, 1108-1110; Hart 1979, 461-465; Neilson, Eckstein, and Hart 1997, 180-182). Maarschalkerweerd divided mounting of people and mounting of other dogs. In the majority of the dogs, mounting of people decreased; 57% of the dogs showed an improvement. When looking at mounting of other dogs, only 40% of the dogs showed a decrease (Maarschalkerweerd et al. 1997, 617-619).

These findings do not correspond with the results of the study performed by Beach. According to his study, surgically castrated dogs do not show less mounting behavior than intact male dogs (Beach 1970, 1-32). This study did show that there were differences in the consummatory phase when comparing castrated dogs to intact dogs. Castrated males established significantly less insertions, and when they did achieve a successful intromission, they needed significantly more mounts to do so (Beach 1970, 1-32). Also, a decrease in the duration of the lock between male and female was seen following castration. One of the castrated dogs even showed total loss of locking behavior. These findings are consistent with the findings of Hart (1968), who also found a significant decrease in the duration of the copulatory lock after castration. Furthermore, Hart's study showed that there is no difference between the effects of surgical castration on sexually experienced castrated males and sexually inexperienced castrated males (Hart 1968, 719-725).

Although there are many studies which indicate that surgical castration leads to a reduction of sexual behavior, there are many individual differences concerning the onset of these changes (Beach 1970, 1-32; Hart 1968, 719-725). While some dogs may show a decrease within 2 weeks after castration, others may continue to mate for a year or even longer. This large individual variation is probably due to environmental and genetic factors (Hart 1974, 383-400).

Besides studies on sexual behavior, much research has been done to determine the effects of castration on roaming behavior in male dogs. Although roaming is not an obvious component of sexual behavior, it is often considered to be a reflection of sexual motivation. Therefore we will give a brief review of what is known about the effects of castration on this behavioral pattern.

In a study performed by Hopkins et al. the behavioral effects of surgical castration were studied in 42 dogs. Roaming proved to be the behavior that was most affected by castration. Over 90 percent of the dogs showed either a rapid or gradual decline in roaming behavior (Hopkins, Schubert, and Hart 1980, 1108-1110). Only one dog did not show an improvement. These findings are rather

consistent to the results of the study performed by Neilson et al. This research showed that 50-70% of adult male dogs show a 50-90% decrease in roaming following castration (Neilson, Eckstein, and Hart 1997, 180-182). In a different study, a reduction of roaming was found in about 90 percent of the cases, with half of the animals showing a rapid decline and the other half showing a gradual decline (Hart 1979, 461-465).

Maarschalkerweerd had a somewhat different approach to investigate the effects of surgical castration on roaming. He split up roaming behavior into two categories: roaming behavior induced by bitches in heat and roaming behavior without an obvious stimulus. Roaming behavior caused by bitches in heat decreased in 57% of the dogs after castration. However, roaming behavior without an obvious stimulus decreased in only 16% of the cases (Maarschalkerweerd et al. 1997, 617-619).

This research does raise some questions. Roaming behavior was split up into roaming behavior caused by bitches in heat and roaming behavior without an obvious stimulus. But what exactly is considered to be an obvious stimulus? The behavioral observations were made by the owners of the dogs. Some things that may not be an 'obvious stimulus' to a dog-owner can be an 'obvious stimulus' for a male dog. For instance: an estrus bitch may leave a scent trail in the environment; this scent will be present long after the bitch has left this area. A dog owner will not consider this to be an 'obvious stimulus' for roaming behavior, since they do not see any female dogs and cannot smell the scent that was left behind. This scent may however very well be the stimulus which causes their dog to display roaming behavior. For this reason, it is doubtful whether the division of roaming behavior into the 2 categories mentioned above can be justified.

Overall, the results which are described above show us that the effects of surgical castration on roaming are dependent of the motivation which causes the objectionable behavior. In many cases roaming is a reflection of sex drive or sexual motivation (Hopkins, Schubert, and Hart 1980, 1108-1110; Hart and Eckstein 1997, 331-344). In these cases surgical castration is an effective method of altering this behavior in the majority of male dogs. If roaming is caused by another stimulus, surgical castration may not be as effective.

Play Behavior

To the best of our knowledge, no literature can be found discussing the effects of surgical castration on play behavior in dogs. However, some articles are available discussing play behavior in rats. In rats, there is a difference in play behavior between male and female rats. Male rats tend to display more play-soliciting behavior than females. Also, females are more likely to withdraw from a play initiation or to withdraw once involved in play (Vanderschuren, Niesink, and Van Ree 1997, 309-326).

Neonatal castration of male rats leads to a reduction of play behavior to the level of females. However, post-weaning castration does not lead to a change in play reaction (Vanderschuren, Niesink, and Van Ree 1997, 309-326; Smith et al. 1996, 215-226). This suggests that the sex-differences in play behavior result from the organizational effects of gonadal hormones in the perinatal period, rather than from the effects of these hormones during puberty.

These findings do not correspond to the results presented in the article of Vinke et al. (2008). In this research, play behavior in ferrets was assessed after (postpubertal) castration. The ferrets

from the placebo group were significantly less playful than the surgically castrated ferrets (Vinke et al. 2008, 104-121).

Fear

Although the role of testosterone and castration status on behaviors such as aggression and sexual behavior has been thoroughly investigated, little is known about its effects on fear responses. Several studies have shown that there are sex differences in fear responses. Females are, overall, more likely to show fear responses than males (Goddard and Beilharz 1983, 299-315). This suggests that the response to fear-related stimuli might be sexually dimorphic and that the hormonal environment may play a role in inducing fear responses.

King et al (2004) investigated the role of testosterone on fear responses in male rats. The aim of this study was to determine whether or not testosterone depletion results in a heightened fear response in rats. The results of this study showed that castrated male rats were significantly more fearful than intact male rats (King, De Oliveira, and Patel 2005, 333-340). Similar results can be found when looking at the effects of orchietomy on fear reactions of male sheep. Castrated rams were significantly more fearful than intact rams in 2 out of 3 test situations. In the third test situation, castrated sheep were also more fearful, though not significantly (Vandenhede and Bouissou 1996, 211-224).

Little research has been done specifically on the effects of orchietomy on fear in dogs. It is often said that dogs become more fearful after castration. We could however, not find any literature to support this hypothesis. However, some articles can be found in which the topic of fearfulness in dogs after castration is addressed. In one of these studies dog owners (n=7) were asked to assess their dog's fear of sounds, people, dogs, traffic and other causes prior to and after castration. This led to the following results: none of the dogs showed an increase of fear following castration, in one dog a decrease in fear of sounds was seen, in another dog, fear of traffic decreased and fear of other causes decreased in 2 dogs. Fear of people and fear of dogs were unaffected by orchietomy (Maarschalkerweerd et al. 1997, 617-619). According to Neilson et al (1997) the probability of seeing a decrease in fear of inanimate stimuli after castration is extremely low (<1%)(Neilson, Eckstein, and Hart 1997, 180-182). In another study which focused on fear related behavior of dogs in the veterinary practice, no significant relation could be found between castration status and fearful behavior (Döring et al. 2009, 38-43).

Behavioral effects of chemical castration

Little is known about the behavioral effects of chemical castration using a GnRH agonist. However, a few studies have been performed.

In a study by Vinke et al. the effects of surgical and chemical castration on inter-male aggression, sexual behavior and play behavior in the male ferret were assessed and compared. For this experiment 21 ferrets, which were kept individually under identical circumstances in the department of clinical sciences of companion animals, were divided into 3 groups: a placebo group, a deslorelin implant group and a castrate group. The 'castrate group' was surgically castrated 7 weeks prior to the start of the behavioral observations and the deslorelin implant group and the placebo group received their implants 7 weeks prior to the start of the behavioral observations.

Three confrontation tests were performed 7 weeks after treatment to evaluate aggression, play behavior and male sexual behavior. All males were confronted with a male from the same test group, with or without the presence of a receptive female (= test 2). During test 3 each male was confronted with a female ferret in estrus, in order to test the sexual motivation of the males. These tests lead to some interesting results. Chemical castration resulted in a decrease of inter-male aggression both in the presence as well as in the absence of a receptive female. Furthermore, chemical castration had a greater effect on the decrease of aggression than surgical castration (Vinke et al. 2008, 104-121).

Secondly, after treatment, sexually motivated behavior was reduced in the deslorelin group as well as in the castrate group during the male-female confrontation. Moreover, this study showed that intact males have a lower incidence of play behavior compared to the deslorelin group, and to a lesser extent the surgically castrated group.

In 2001 a study was performed to determine the effects of deslorelin treatment on the control of reproduction and sex related behavior in exotic wild animals (Bertschinger et al. 2001, 275-283). Although this study was very brief and small numbers of animals were involved, the following preliminary observations were made: antagonistic behavior in male sea otters ceased after treatment with a deslorelin implant (6mg) and no adverse effects were observed on social behavior in four male cheetahs treated with deslorelin. In seven red and five grey male wolves, treatment was ineffective at controlling spermatogenesis as well as at controlling sexual behavior.

In dogs, little research has been done to determine the behavioral effects of chemical castration. In 2010 an article was published which revealed the outcome of a study that was designed to determine the effect of the slow release GnRH agonist azagly-nafarelin (Gonazon ®), on benign prostatic hyperplasia, hyper-sexuality, aggressive behavior and excessive micturition (Goericke-Pesch et al. 2010, 920-926). The behavioral changes were assessed by the owners of the dogs by scoring the changes as "significant improvement", "partial improvement" or "no improvement". Territorial micturition ceased in all dogs (n=2) which showed this problem behavior. Hypersexuality decreased in all dogs (n= 26), with a significant improvement in 24 dogs. Of the dogs showing aggressive behavior, 75% showed an improvement. This improvement was significant in 15 dogs (47%). Overall, this study shows that Gonazon is efficient in decreasing hypersexuality, as clinical signs improved in practically all dogs. Also, aggressiveness clearly decreased and this decrease is

comparable to the improvement of aggressiveness after surgical castration as was previously shown (Goericke-Pesch et al. 2010, 920-926). It must however be taken into consideration that the observations that were made, were subjective observations, since they were made by the dog owners. Little can be said about the effects of Gonazon treatment on excessive micturition, since this behavior was only observed in two animals.

Aim of the study

The goal of this study is to determine the effects of 'chemical castration' with a suprelorin implant on the behavior of intact male dogs and to determine whether or not these effects are comparable to the behavioral effects of surgical castration. Behavioral tests as well as questionnaires were used to evaluate the behavioral changes in the privately owned dogs. Several behavioral elements have been assessed. Given the extensiveness of these tests, we will however focus on 4 types of behavior in this report: Aggression, Sexual Behavior, Play Behavior and Fear/Insecurity.

Materials and Methods

Selection of dogs - dogs included in this study were intact adult males of various breeds and ages, but with a minimum age of 18 months. Furthermore, the dogs needed to have two scrotal testicles and no general health problems in order to be included in the study.

This study was designed as a prospective, non blinded, non randomized paired study.

In total, we collected a complete data set of 41 dogs, 23 of these dogs were chemically castrated with a 4.7 mg Suprelorin implant at the University clinic for Companion Animals in Utrecht and 18 dogs were surgically castrated either at the University clinic for Companion Animals in Utrecht or by their own veterinarian.

Data Collection – data were collected in two different ways shortly before and 4-5 months after treatment; owners were asked to fill out a questionnaire and a behavior test was performed.

Questionnaire: The dog owners were asked to fill out a questionnaire at two different times: one prior to treatment (T=0), and one approximately four to five months after treatment (T=2). The questionnaire consisted of different types of questions designed to collect information about the environment of the dog, the motivation for castrating the dog, the dog's general health, and different types of behavior.

The first questionnaire consisted of 33 questions. In the second questionnaire 5 extra questions were added, these questions were perceptive questions designed to assess whether the owner felt that the dog's behavior had changed after treatment, and if so, to what extent.

One of the questions which was present in both questionnaires (question 9) was designed to ask the owner how his or her dog responds to certain stimuli. Multiple responses could be given to this question.

The complete questionnaire can be found in appendix 1.

Behavior Test:

The behavior test was performed to acquire an objective picture of the dog's behavior. The behavior test was performed at two times; prior to treatment (T=0) and four to five months after treatment (T=2). The behavior test consisted of 18 different tests, a description of these tests can be found in appendix 2.

The behavior tests were registered with a video camera so scoring of the observed behavior could be performed at a later date. The following components were scored (if applicable): approach, sociability, play behavior, fright, recovery after fright, fear/insecurity, submissive/dominant behavior, level of activity, aggressiveness, posture, exploratory behavior, pulling on the leash, obedience, sexual behavior, reaction to restraint and the reaction to being stared at.

Scoring of the behavior tests

The different behavioral components that could be observed during the behavior tests have been scored using the ethogram provided in appendix 3. To eliminate inter-observer variation, all videos were scored by one person. The veterinary student who scored the videos spent several days practicing scoring the behavioral components during which she received extensive guiding from a

behavioral specialist (C.M.Vinke), before starting the definitive scoring of the videos. The order in which the different videos were scored was not defined prior to scoring and was not randomized. It is however important to mention that the 2 videos that were available of each dog were not scored directly after one another, but the video of T= 0 was always scored first. The scoring was non-blinded: the student who scored the tests was aware of which group each dog belonged to.

Statistical Analyses

In order to determine the significance of our results, several statistical analyses were performed.

Logistical Regression:

In order to compare the results of the behavior test at T=0 to the results at T=2 within groups a logistical regression analysis was performed. This test was also used to compare the overall results of the behavioral test at T=0 between groups.

In order to perform this test, the different scoring categories had to be lumped together, so only two categories remained: presence of the behavior in question or absence of the behavior in question.

Chi-square test

Besides looking at the entire group, we also assessed the results of the behavior test of each dog individually in order to determine if the dog showed an increase, a decrease or showed no change in the amount of times a certain behavior was observed. In order to determine if there was a significant difference in these results between the Suprelorin implant group and the surgical castration group, a chi-square test was performed.

The chi-square test was also performed to compare the differences between groups concerning the answers that were given to the perceptive questions in the second questionnaire and to compare the answers given to question 25 at T=0 between groups.

McNemar's Chi-square test

The McNemar's Chi-square test was used to compare the individual results of the behavioral test (increase/decrease/no change) to the answers of the perceptive questions in the second questionnaire within groups.

Also, this test was used to compare the answers to question 25 of the questionnaire at T=0 and T=2 within groups.

All statistical analyses were performed using the software program SPSS 16.0 for Windows. Results were considered significant if the p-value was < 0.05.

Results

Aggression

Suprelorin Implant Group

Behavior Test

The observations that were made in the Suprelorin implant group concerning aggression during T=0 and T=2 are provided in table 1.

At T=0, 4 dogs showed aggression. All cases of aggression were seen during the same test; test 13: meeting an unfamiliar child (doll). Remarkably, all of the dogs that showed aggression also showed a fear/insecurity response during the event in which they showed aggression (score 1). At T=2, 3 dogs showed aggression during one or multiple tests. Aggression was seen during test 6, 9, 10, 13 and 15 and 18. In six out of eight situations in which aggressive behavior was observed, we also saw fear or insecurity. Overall the mean score for aggression at T=0 was 3.99 and at T=2 the mean score was 3.97. Moreover, the number of aggressive reactions observed during the behavioral test did not change following treatment ($p= 0.263$).

If we look at all dogs individually, we can see that 3 dogs showed an increase in the number of situations in which they showed aggression, 2 dogs showed a decrease and in 18 dogs, no change was observed.

Questionnaire

The data concerning aggression which we obtained from question nine are displayed in table 2. If we look at these data, we can see that there is a large percentage of dogs which show aggression towards other intact male dogs (T=0). At T=2 this percentage has decreased from 26.1% to 13.0%.

An important question asked in the second questionnaire (T=2) was the following perceptive question: "*Has your dog's aggression towards other male dogs decreased and if so, to what extent?*" Five out of 23 owners (21.7%) noticed an increase in aggression towards other male dogs. Ten owners (43.5%) noticed a decrease. Of these owners five owners noticed a decrease of <50%, three owners saw a decrease of >50% and two owners saw a decrease of >90%. The remaining eight owners (34.8%) did not notice a change in the amount of aggression towards other male dogs.

Suprelorin Implant n=23	0	1	2	3	4
T=0 (n=624)	0 (0%)	0 (0%)	3 (0.5%)	1 (0.1%)	620(99.4%)
T=2 (n=631)	0 (0%)	0 (0%)	8 (1.3%)	0 (0%)	623(98.7%)

Table 1: Aggressiveness Suprelorin Implant Group

Question 9: Aggression Suprelorin Implant Group	Aggression T= 0 (N)	Aggression T=0 (%)	Aggression T= 2 (N)	Aggression T=2 (%)
9a: Owner	0	0%	0	0%
9B: family members	0	0%	0	0%
9C: familiar children	0	0%	0	0%
9D:unfamiliar man	1	4.3 %	0	0 %
9E:unfamiliar woman	0	0%	0	0 %
9F:unfamiliar child	2	8.7%	0	0%
9G: intact male dog	6	26.1%	3	13.0%
9H: castrated male dog	3	13.0%	2	8.7%
9 I: anestrus bitch	0	0%	1	4.3%
9 J: bitch in heat	1	4.3%	1	4.3%

Table 2: Results Question 9: Aggression Suprelorin Implant group

Surgical Castration n= 18	0	1	2	3	4
T=0 (n=475)	0 (0%)	4 (0.8%)	7 (1.5%)	0 (0%)	464(97.7%)
T=2 (n=483)	0 (0%)	6 (1.2%)	5 (1.0%)	0 (0%)	472(97.7%)

Table 3: Aggressiveness Surgical Castration Group

Question 9: Aggression Surgical Castration Group	Aggression T= 0 (N)	Aggression T=0 (%)	Aggression T= 2 (N)	Aggression T=2 (%)
9a: Owner	1	5.6%	0	0%
9B: family members	1	5.6%	1	5.6%
9C: familiar children	0	0%	1	5.6%
9D:unfamiliar man	4	22.2%	3	16.7%
9E:unfamiliar woman	4	22.2%	2	11.1%
9F:unfamiliar child	4	22.2%	2	11.1%
9G: intact male dog	7	38.9%	5	27.8%
9H: castrated male dog	4	22.2%	5	27.8%
9 I: anestrus bitch	2	11.1%	4	22.2%
9 J: bitch in heat	2	11.1%	2	11.1%

Table 4: Results Question 9: Aggression Surgical Castration Group

Surgical Castration Group

Behavior Test

The observations that were made in the surgical castration group concerning aggression during T=0 and T=2 are provided in table 3.

At T=0, 8 dogs showed aggression in one or multiple situations. Aggression was seen during test 1, 11, 13 and 15. In 8 out of 11 situations (72.7 %) in which aggressive behavior was observed, we also saw fear or insecurity. At T=2, 4 dogs showed aggression during one or multiple tests. Aggression was seen during test 1, 2, 11, 13, 15, 16, 17 and 18. In 4 out of 11 situations (36.4 %) in which aggressive behavior was observed, we also saw fear or insecurity.

Overall the mean score for aggression at T=0 was 3.95 and at T=2 the mean score was 3.94. Additionally, the number of aggressive reactions observed during the behavioral test did not change following treatment ($p= 0.968$).

If we look at all dogs individually, we see a decrease in the amount of aggressive behavior in 5 dogs, an increase in 4 dogs and no change could be found in the remaining 9 dogs.

Questionnaire

Table 4 shows the results of question 9 at T=0 and T=2.

A decrease in aggressive behavior after treatment occurred in five situations and an increase is seen in 3 situations. Differences are however small.

The owners of the dogs in the surgical castration group were also asked the following perceptive question: *"Has your dog's aggression towards other male dogs decreased and if so, to what extent?"* Two out of 17 owners (11.8%) noticed an increase in the amount of aggression shown by there dogs in reaction to other male dogs. Eight owners (47.0%) noticed a decrease, of these owners 2 owners noticed a decrease of <50%, six owners noticed a decrease of >50% and none of the owners saw a decrease of >90%. The remaining 7 owners (41.2%) did not see any change.

Suprelorin Implant Group vs. Surgical Castration Group

The changes that were seen in the behavioral test as well as the questionnaire concerning aggression following treatment in the Suprelorin implant group and the surgical castration group are reflected in figure 1.

Behavior Test

At T=0 the dogs in the Suprelorin implant group showed slightly less aggressive behavior based on the overall means, compared to the dogs in the surgical castration group (3.99 and 3.95 respectively, 4= no observed aggression). If we however compare the total number of aggressive behaviors observed in the suprelorin implant group and the surgical castration group at T=0 (4 and 11 respectively) we do find a significant difference ($p=0.027$); more cases of aggressive behavior were seen in the surgical castration group. Furthermore no difference between groups was observed concerning change in aggressive behavior during the studied period (increase, decrease and no change: $p= 0.679$, $p=0.209$ and $p= 0.058$ respectively; table 5)

Questionnaire

If we look at the answers to the perceptive question concerning aggression in the questionnaire, we find that there are not a lot of differences between the answers given by the owners of the dogs of the Suprelorin implant group and the owners of the dogs of the surgical castration group. There are no significant differences between the number of owners that noticed a decrease, an increase or no change in aggression towards other male dogs ($p= 0.822$, $p= 0.677$ and $p=0.680$ respectively).

<i>Change in Aggression</i>	Increase	Decrease	No change
Suprelorin Implant Group (n=23)	13.0%	8.7%	78.3%
Surgical castration Group (n=18)	27.8%	22.2%	50.0%

Table 5: Changes in Aggressive Behavior during behavioral test

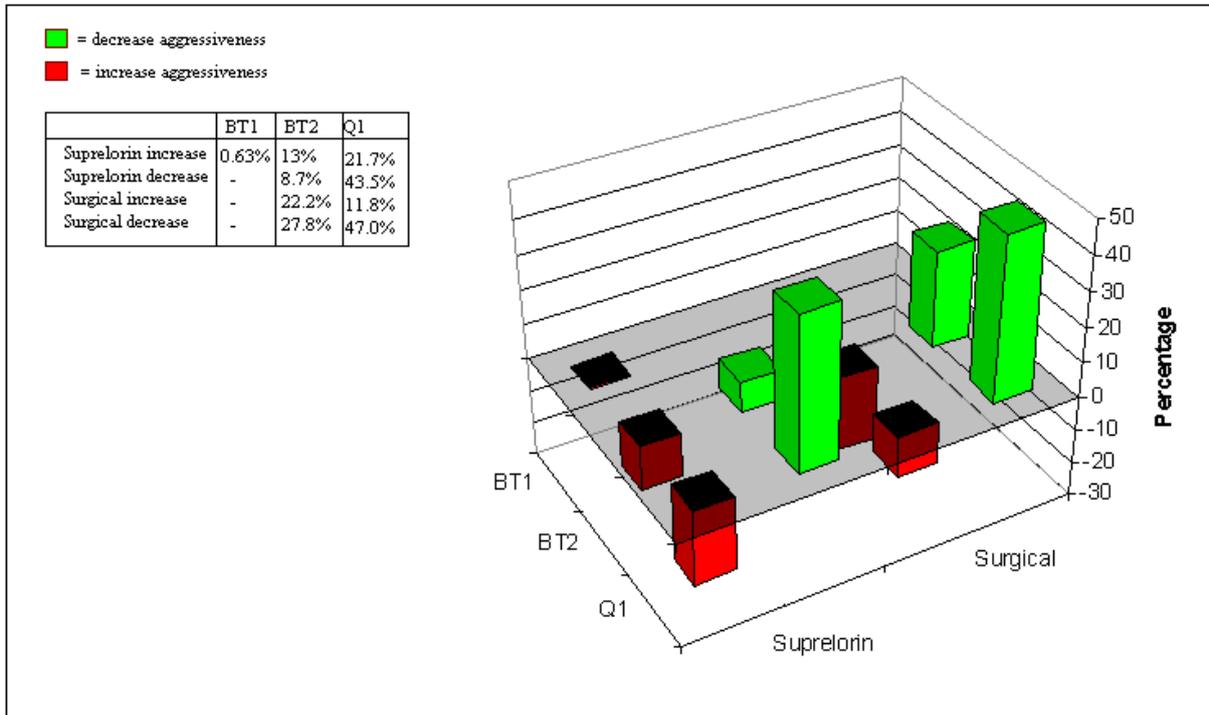


Figure 1: Changes in Aggressive Behavior

BT1: Behavioral Test Entire group. Difference between percentage of aggressiveness during T=0 and T=2.

BT2: Individual Results of behavioral test. Percentage of dogs that show an increase in aggressive behavior and percentage of dogs that show a decrease in aggressive behavior.

Q1: Responses to perceptive question. Percentage of owners that noticed an increase in aggressive behavior towards other male dogs and percentage of owners that noticed a decrease.

Sexual Behavior

Suprelorin Implant Group

Behavior Test

The observations made during the behavioral test concerning sexual behavior at T=0 and T=2 are presented in table 6.

Suprelorin Implant n=23	Score = 0	Score =1	Score =3	Score =4
T=0 (n=115)	0	18 (15.7%)	21 (18.3%)	76 (66.1%)
T=2 (n=115)	0	1 (0.9%)	26 (22.6%)	88 (76.5 %)

Table 6: Sexual Behavior Suprelorin Group

Overall the mean score for sexual behavior of the suprelorin implant group at T=0 was 3.35 ±1.08 and the mean score at T=2 was 3.75 ± 0.49. Additionally, if we compare the total number of situations during which sexual behavior was observed at T=0 and T=2, we find that there has been a decrease with a tendency towards statistical significance (p=0.082). If we assess the individual results of each dog of Test 18 (Meeting a bitch in heat) at T=0 and T=2 we find that 15 dogs (65.2%) showed a decrease in sexual behavior, 2 dogs (8.7%) showed an increase in sexual behavior and 6 dogs (26.1%) showed no change.

Questionnaire

Several questions concerning sexual behavior were asked in the questionnaire. One of these questions was: "25. Does your dog ever show any sexual behavior?"

The owners were asked to answer this question in the first questionnaire prior to treatment (T=0) as well as in the second questionnaire which was filled out after treatment (T=2). At T=0, 18 owners (78.3%) answered 'yes', while at T=2 only 8 owners (34.8%) answered 'yes'. This is a significant decrease (p = 0.006).

In the second questionnaire the following perceptive question was asked: "Has your dog's sexual behavior towards estrus bitches decreased and if so, to what extent?" Eight out of 21 owners (38.1%) noticed a decrease in their dog's sexual behavior towards estrus bitches. Of these owners, one owner noticed a decrease of <50%, five owners noticed a decrease of >50% and 2 owners noticed a decrease of >90%. The remaining thirteen owners (61.9%) did not notice any change.

Behavior Test vs. Questionnaire

If we compare the results of behavioral test 18 to the answers to the perceptive question in the second questionnaire, we find that the decrease observed during the behavioral test is significantly greater than the decrease observed by the owners (p = 0.016). Also, the percentage of owners that did not notice any change in their dog's behavior is significantly greater than the percentage of dogs that did not show any change during the behavioral test (p =0.016). There is no significant difference between the number of dogs that showed an increase in sexual behavior during the behavioral test and the number of owners that noticed an increase (p = 0.500).

Surgical Castration Group

Behavior Test

The results of the behavioral test at T=0 and T=2 of the surgical castration group concerning sexual behavior are presented in table 7.

Surgical Castration N=18	Score = 0	Score =1	Score =3	Score =4
T=0 (n=88)	1 (1.1%)	11 (12.5%)	9 (10.3%)	67 (76.1%)
T=2 (n= 86)	1 (1.2%)	2 (2.3%)	12 (14.0%)	71 (82.6%)

Table 7: Sexual Behavior Surgical Castration Group

Overall the mean score for sexual behavior of the surgical castration group at T=0 was 3.48 ± 1.07 and the mean score at T=2 was 3.74 ± 0.69 . In addition, the total number of sexual reactions observed during the behavioral test did not change after treatment ($p=0.297$).

If we assess the individual results of each dog of Test 18 (Meeting a bitch in heat) at T=0 and T=2, we find that 8 dogs (44.4%) showed a decrease in sexual behavior, 2 dogs (11.1%) showed an increase in sexual behavior and 8 dogs (44.4%) showed no change.

Questionnaire

Several questions concerning sexual behavior were asked in the questionnaire. One of these questions was: "25. Does your dog ever show any sexual behavior?"

The owners were asked to answer this question in the first questionnaire prior to treatment (T=0) as well as in the second questionnaire which was filled out after treatment (T=2). At T=0, 16 owners (88.9%) answered 'yes', while at T=2 only 6 owners (33.3%) answered 'yes'. This is a significant decrease ($p = 0.002$).

In the second questionnaire the following perceptive question was asked: "Has your dog's sexual behavior towards estrus bitches decreased and if so, to what extent?"

13 out of 17 owners (76.5%) noticed a decrease in their dog's sexual behavior towards estrus bitches. Of these owners, five owners noticed a decrease of >50% and eight owners noticed a decrease of >90%. The remaining four owners (23.5%) did not notice any change.

Behavior Test vs. Questionnaire

If we compare the results of behavior test 18 to the answers to the perceptive question in the second questionnaire, we find that there are no significant differences between the amount of dogs in which an increase, a decrease or no change was observed ($p = 0.500$, $p = 0.062$ and $p = 0.125$ respectively).

Suprelorin Implant Group vs. Surgical Castration Group

The changes that were seen in the behavior test as well as the questionnaire concerning sexual behavior following treatment in the Suprelorin implant group and the surgical castration group are reflected in figure 2.

Behavior Test

If we compare the results of the Suprelorin implant group to the results of the surgical implant group at T=0, we do not find a significant difference between the amount of sexual behavior displayed by the dogs ($p = 0.121$).

By looking at the individual results of test18 from the dogs from both groups, we do not find any significant differences between groups concerning the amount of dogs in which an increase, a decrease or no change was observed ($p = 1.000$, $p = 0.183$ and $p = 0.219$ respectively, table 8).

Questionnaire

If we look at the answers given to question 25 of the questionnaire, we can see that at T=0 the percentage of dogs who show sexual behavior is greater in the surgical castration group than in the suprelorin implant group (88.9% and 78.3% respectively). This difference is not significant ($p = 0.438$). In both groups a decrease in this percentage is seen in the questionnaire at T=2. This decrease is greater for the surgical castration group (T=0:88.9%, T=2:33.3%) than in the suprelorin implant group (T=0: 78.3%, T=2:34.8%).

If we look at the answers to the perceptive question concerning sexual behavior in the questionnaire, we find that there are big differences between the answers given by the owners of the dogs of the suprelorin implant group and the owners of the dogs of the surgical castration group. Significantly more owners of the surgical castration group noticed a change in their dog's behavior following treatment compared to the owners of the suprelorin implant dogs ($p=0.018$). Also, significantly more owners from the surgical castration group noticed a decrease in the sexual behavior towards estrus bitches compared to the owners of the suprelorin implant group ($p = 0.018$). None of the owners reported an increase of sexual behavior.

<i>Change in Sexual behavior</i>	Increase	Decrease	No change
Suprelorin Implant Group (n=23)	8.7%	65.2%	26.1%
Surigcal castration Group (n=18)	11.1%	44.4%	44.4%

Table 8: Changes in Sexual Behavior during Behavioral Test

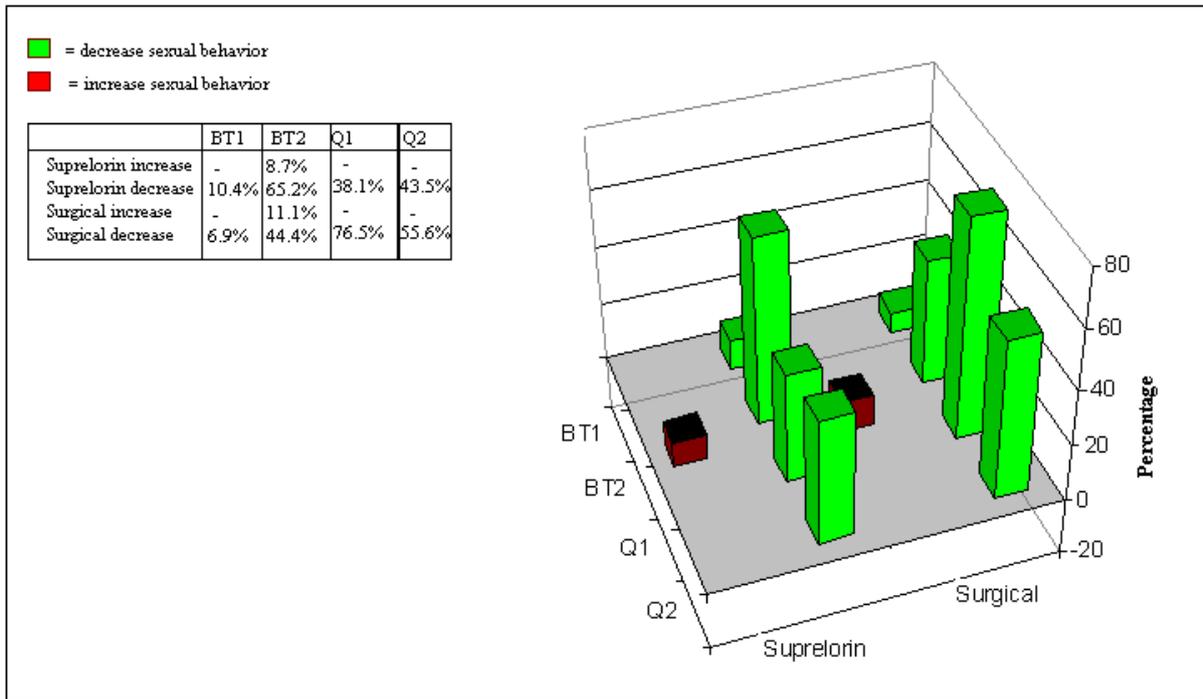


Figure 2: Changes in Sexual Behavior

BT1: Behavioral Test Entire group. Difference between percentage of sexual behavior during T=0 and T=2.

BT2: Individual Results of behavioral test. Percentage of dogs that show an increase in sexual behavior and percentage of dogs that show a decrease in sexual behavior when meeting an estrus bitch (test18).

Q1: Responses to perceptive question. Percentage of owners that noticed an increase in sexual behavior towards bitches in heat and percentage of owners that noticed a decrease.

Q2: Difference (percentage) between the answers given to question 25 At T=0 and T=2.

Play Behavior

Suprelorin Implant group

Behavior Test

The observations that were made in the Suprelorin implant group concerning play behavior during T=0 and T=2 are provided in table 9.

In the behavioral test the overall mean score of play behavior of the Suprelorin implant group at T=0 and T=2 were not similar (0.27 ± 0.80 and 0.44 ± 1.04 respectively). In addition, the number of play reactions observed during the behavior test increased after treatment ($p=0.008$).

If we look at the results of all dogs individually, we find that 3 dogs (13%) showed a decrease in the amount of play behavior during the behavioral test, 13 dogs (56.5%) showed an increase and in 7 dogs (30.4%) no change in the number of play reactions was found.

Questionnaire

The results of question nine concerning playful behavior are provided in table 10.

These data show us that in half of the presented situations, more dogs reacted in a playful manner when confronted with the stimulus after treatment compared to the number of dogs that reacted playful before treatment. There are however, also three situations in which the number of dogs that showed play behavior decreased.

The owners of the dogs in the Suprelorin implant group were asked to answer the following question in the second questionnaire (T=2): "*Has the amount of play behavior of your dog changed?*" Three out of 23 owners (13%) reported a decrease in the amount of play behavior shown by their dog since the treatment; 10 owners (43.5%) noticed an increase in play behavior and 10 owners (43.5%) did not see any change in the play behavior displayed by their dog. Of the 10 owners that noticed an increase in play behavior, 5 owners appointed the increase as a <50% increase and 5 owners saw a >50% increase.

Behavior Test vs. Questionnaire

If we compare the results of the behavior test to the results of the questionnaire, we find that there is no significant difference between the behavior test and the questionnaire in any of the three categories: decrease, increase or no change ($p = 1.000$, $p = 0.250$ and $p = 0.250$ respectively).

Suprelorin Implant n=23	Score = 0	Score = 1	Score =3	Score =4
T=0 (n=625)	539 (86.2%)	47 (7.5%)	32 (5.1%)	7 (1.1%)
T=2 (n=631)	509 (80.7%)	52 (8.2%)	52 (8.2%)	18 (2.9%)

Table 9: Play Behavior Suprelorin Implant Group

Question 9: Play Behavior Suprelorin Implant Group	Play Behavior T= 0 (N)	Play Behavior T=0 (%)	Play Behavior T= 2 (N)	Play Behavior T=2 (%)
9a: Owner	15	65.2%	15	65.2%
9B: family members	11	47.8%	14	60.9%
9C: familiar children	9	39.1%	9	39.1%
9D:unfamiliar man	4	17.4%	8	34.8%
9E:unfamiliar woman	5	21.7%	7	30.4%
9F:unfamiliar child	8	34.8%	9	39.1%
9G: intact male dog	4	17.4%	3	13.0%
9H: castrated male dog	6	26.1%	4	17.4%
9 I: bitch which is not in heat	12	52.2%	8	34.8%
9 J: bitch in heat	2	8.7 %	4	17.4 %

Table 10: Results Question 9: Play Behavior Suprelorin Implant Group

Surgical Castration n=18	Score = 0	Score =1	Score =3	Score =4
T=0 (n=475)	426 (89.7%)	26 (5.5%)	16 (3.4%)	7 (1.5%)
T=2 (n=483)	412 (85.3%)	36 (7.5%)	23 (4.8%)	12 (2.5%)

Table 11: Play Behavior Surgical Castration Group

Question 9: Play behavior Surgical Castration Group	Play Behavior T= 0 (N)	Play Behavior T=0 (%)	Play Behavior T= 2 (N)	Play Behavior T=2 (%)
9a: Owner	12	66.7%	15	83.3%
9B: family members	11	61.1%	14	77.8%
9C: familiar children	5	27.8%	8	44.4%
9D:unfamiliar man	4	22.2%	6	33.3%
9E:unfamiliar woman	4	22.2%	5	27.8%
9F:unfamiliar child	6	33.3%	5	27.8%
9G: intact male dog	4	22.2%	4	22.2%
9H: castrated male dog	6	33.3%	7	44.4%
9 I: bitch which is not in heat	11	61.1%	9	50.0%
9 J: bitch in heat	7	38.9%	8	44.4%

Table 12: Results Question 9: Play Behavior Surgical Castration Group

Surgical Castration Group

Behavior Test

The observations that were made in the surgical castration group concerning play behavior during T=0 and T=2 are provided in table 11.

In the behavior test the overall mean score of play behavior of the surgical castration group at T=0 and T=2 were not similar (0.21 ± 0.74 and 0.32 ± 0.90 respectively). In addition, the number of playful reactions observed during the behavior test increased after treatment ($p=0.041$).

If we look at the data of the behavior tests of each dog individually, we find that 4 dogs (22.2%) showed a decrease in play behavior, 10 dogs (55.6%) showed an increase in play behavior, and in 4 dogs (22.2%) no change was observed.

Questionnaire

Table 12 shows an overview of the responses to question 9 in the surgical castration group. In 7 out of 10 cases, the number of dogs which reacted playful to the stimulus increased after castration. In two cases we can see a decrease.

The owners of the dogs in the surgical castration group were asked to answer the following perceptive question in the second questionnaire (T=2): "*Has the amount of play behavior of your dog changed?*" Nine out of 18 owners (50%) noticed an increase in play behavior since their dog had been castrated, the other 50% of the owners did not notice any change. Four owners noticed an increase of <50%, another four owners noticed an increase of >50%. The remaining owner noticed an increase of >90%.

Behavior Test vs. Questionnaire

If we statistically compare the three different categories of change (decrease, increase, no change) of the behavior test and the questionnaire to each other, we do not find any significant differences ($p = 0.125$, $p = 1.000$ and $p = 0.062$ respectively).

Suprelorin Implant Group vs. Surgical Castration Group

The changes that were seen in the behavior test as well as the questionnaire concerning play behavior following treatment in the Suprelorin implant group and the surgical castration group are reflected in figure 3.

Behavior Test

At T=0 no difference in play behavior was observed in the behavior test between both treatment groups ($p=0.09$). Furthermore no difference between groups was observed concerning change in play behavior during the studied period (increase, decrease and no change: $p= 0.890$, $p=0.679$ and $p= 0.726$ respectively; table 13)

Questionnaire

If we look at the answers to the perceptive question concerning play behavior in the questionnaire, we find that there are not a lot of differences between the answers given by the owners of the dogs of the Suprelorin implant group and the owners of the dogs of the surgical castration group. There are no significant differences between the number of owners that noticed a decrease, an increase or no change in play behavior ($p= 0.243$, $p= 0.678$ and $p=0.678$ respectively).

<i>Change in Play Behavior</i>	Increase	Decrease	No change
Suprelorin Implant Group	56.5%	13.0%	30.5%
Surgical castration Group	55.6%	22.2%	22.2%

Table 13: Changes in Play Behavior during Behavioral Test

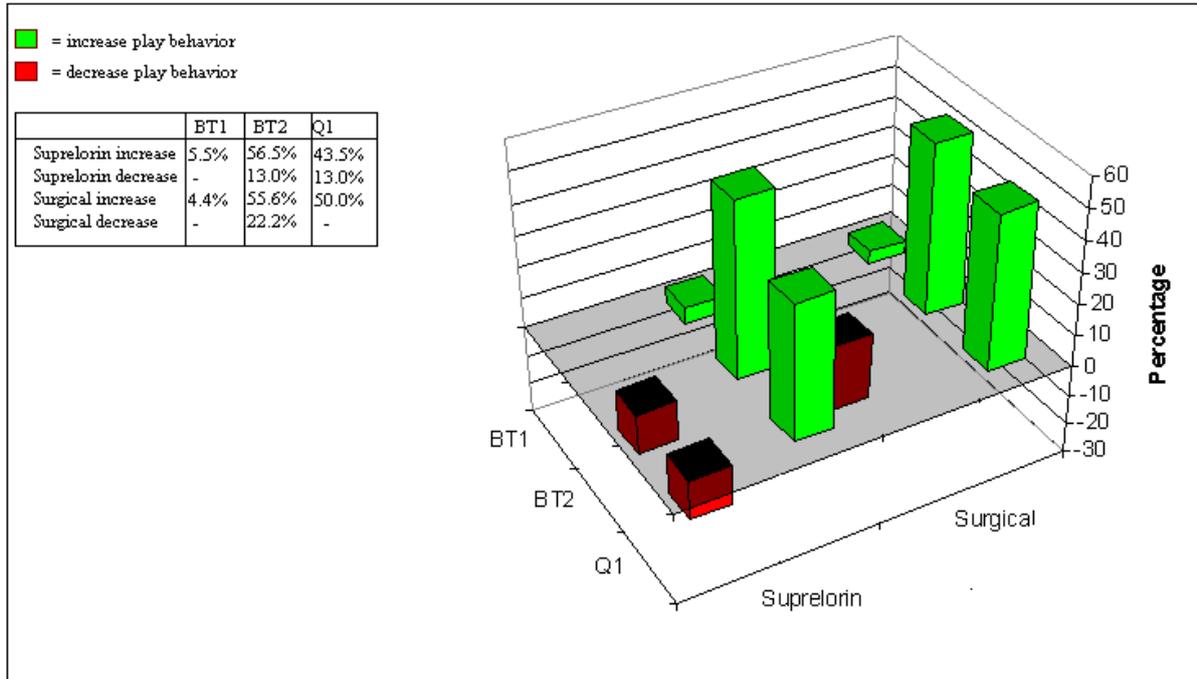


Figure 3: Changes in Play Behavior

BT1: Behavioral Test Entire group. Difference between percentage of play behavior during T=0 and T=2.

BT2: Individual Results of behavioral test. Percentage of dogs that show an increase in play behavior and percentage of dogs that show a decrease in play behavior.

Q1: Responses to perceptive question. Percentage of owners that noticed an increase in play behavior and percentage of owners that noticed a decrease.

Fear/insecurity

Suprelorin Implant group

Behavior test: the observations that were made in the Suprelorin implant group concerning fear/insecurity at T=0 and T=2 are provided in table 14.

In the behavior test the overall mean score of fear/uncertainty of the Suprelorin group at T=0 and T=2 were similar (3.67 ± 0.66 and 3.70 ± 0.65 respectively). In addition, the number of fear reactions that were observed also did not change after treatment ($p=0.28$). If we compare the results of the behavioral tests at T=0 and T=2 of all the dogs individually we find that 13 dogs (56.5%) showed a decrease, six dogs (26.1%) showed an increase, and four dogs (17.4%) showed no change in the number of fear or uncertainty reactions observed during the test.

Questionnaire

The responses to question nine concerning fear/insecurity can be found in table 15. These data show that the number of dogs that show fear in response to unfamiliar men, intact male dogs and castrated male dogs have increased after treatment. The number of dogs which show fear/uncertainty as a reaction to familiar children, unfamiliar women and anestrus bitches decreased after treatment. However, differences are small.

One of the most important questions concerning fear/uncertainty is the perceptive question asked in the second questionnaire (T=2): "*Do you think your dog has become more fearful or insecure after the treatment?*" 17 out of 23 owners (74%) noticed no change of fear or uncertainty in their dogs, the remaining 6 (26%) owners noticed an increase. Of these owners, three owners noticed an increase of less than 50%, two owners said the increase was more than 50% and one owner said that the increase was more than 90%.

Behavior Test vs. Questionnaire

Comparing the results of the perceptive question to the behavior test a larger number of dogs showed a decrease of fear or uncertainty ($p = 0.000$) during the behavior test. There is no significant difference between the questionnaire and the behavior test concerning the number of dogs that showed an increase in fear/uncertainty ($p = 1.000$). A significant difference was however found if we compare the number of dogs that showed no change concerning fear/uncertainty. The number of dogs that showed no change was significantly greater in the questionnaire ($p = 0.000$).

Suprelorin Implant (n=23)	Score: 0	Score: 1	Score: 3	Score: 4
T=0 (n=625)	0 (0%)	22 (3.5%)	141 (22.6%)	462 (73.9%)
T=2 (n=631)	1 (0.2%)	20 (3.2%)	127 (20.1%)	483 (76.5%)

Table 14: Fear/insecurity Suprelorin Implant Group

Question 9: Fear/Insecurity Suprelorin Implant Group	Fear/Insecurity T= 0 (N)	Fear/Insecurity T=0 (%)	Fear/Insecurity T= 2 (N)	Fear/Insecurity T=2 (%)
9a: Owner	0	0%	0	0%
9B: family members	0	0%	0	0%
9C: familiar children	2	7%	0	0%
9D: unfamiliar man	3	13%	4	17%
9E: unfamiliar woman	2	9%	1	4%
9F: unfamiliar child	3	13%	3	13%
9G: intact male dog	5	22%	7	30%
9H: castrated male dog	2	9%	3	13%
9 I: anestrus bitch	1	4%	0	0%
9 J: bitch in heat	0	0%	0	0%

Table 15: Results question 9: Fear/Insecurity Suprelorin Implant Group

Surgical Castration N=18	Score = 0	Score = 1	Score = 3	Score = 4
T=0 (n=475)	0 (0%)	14 (2.9%)	98 (20.6%)	363 (76.4%)
T=2 (n=483)	0 (0%)	15 (3.1%)	100 (20.7%)	368 (76.2%)

Table 16: Fear/Insecurity Surgical Castration Group

Question 9: Fear/Insecurity Surgical Castration Group	Fear/Insecurity T= 0 (N)	Fear/Insecurity T=0 (%)	Fear/Insecurity T= 2 (N)	Fear/Insecurity T=2 (%)
9a: Owner	0	0%	0	0%
9B: family members	1	5.6%	1	5.6%
9C: familiar children	1	5.6%	2	11.1%
9D: unfamiliar man	8	44.4%	6	33.3%
9E: unfamiliar woman	9	50%	7	38.9%
9F: unfamiliar child	4	22.2%	5	27.8%
9G: intact male dog	4	22.2%	4	22.2%
9H: castrated male dog	4	22.2%	5	27.8%
9 I: anestrus bitch	3	16.7%	4	22.2%
9 J: bitch in heat	1	5.6%	3	16.7%

Table 17: Results Question 9: Fear/Insecurity Surgical Castration Group

Surgical Castration Group:

Behavior Test:

The observations that were made in the surgical castration group concerning fear/insecurity during T=0 and T=2 are provided in table 16.

In the behavior test the overall mean score of fear/uncertainty of the surgical castration group at T=0 and T=2 were similar (3.71 ± 0.62 and 3.70 ± 0.63 respectively). In addition, the number of fear reactions observed during the behavior tests also did not change after treatment ($p=0.933$).

If we compare the results of the behavior tests at T=0 and T=2 of all the dogs individually we find that 8 dogs (44.4%) showed a decrease, five dogs (27.8%) showed an increase, and five dogs (27.8%) showed no change in the number of fear or uncertainty reactions observed during the test .

Questionnaire:

In table 17 we can see the responses to question 9 concerning fear/uncertainty of the owners of the surgical castration dogs. If we compare the number of dogs that show fear/uncertainty following a certain stimulus at T=0 to the number of dogs that show this reaction at T=2, we find that a decrease has taken place in 2 out of 10 situations. We also see that an increase of fear/uncertainty is present in 5 out of 10 situations.

Similar to the owners of the suprelorin castration group, the owners of the dogs of the surgical castration group were also asked to answer the following question after treatment (T=2): "*Do you think your dog has become more fearful or uncertain after the treatment?*" 14 out of 18 owners (77.8%) noticed no change of fear or insecurity in their dogs, two owners (11.1%) noticed an increase, one of <50% and one of >50%. The remaining two owners (11.1%) noticed a decrease.

Behavior Test vs. Questionnaire:

If we compare the results of the behavior test to the results of the perceptive question we find that significantly more decreases of fear/uncertainty were seen during the behavior test ($p = 0.031$). If we compare the number of dogs that showed no change in behavior during the behavior test to the number of dogs that showed no change according to their owner, we find that there is a significant difference. The number of dogs that did not show a change of behavior was greater in the questionnaire ($p = 0.004$). No significant difference was found concerning increases of fear/insecurity ($p = 0.250$).

Suprelorin Implant group vs. Surgical Castration group

The changes that were seen in the behavior test as well as the questionnaire concerning fear/insecurity following treatment in both test groups are reflected in figure 4.

Behavior test:

At T=0 no difference in fear or uncertainty was observed in the behavior test between both treatment groups ($p=0.343$). Furthermore no difference between groups was observed concerning change in fear or uncertainty during the studied period (increase, decrease and no change: $p=1.00$, $p=0.443$ and $p= 0.471$ respectively; table 18)

Questionnaire:

If we look at the answer to the perceptive question, we can see some differences between the Suprelorin implant group and the surgical castration group. Two owners of the surgical castration group noticed a decrease of fear/uncertainty in their dog, while in the Suprelorin implant group none of the owners noticed a decrease. This difference is however not significant ($p = 0.187$).

Also, a greater percentage of the owners of the Suprelorin implant group noticed an increase in fear/uncertainty than the owners of the surgical castrate group (26.1% and 11.1% respectively). This difference is also not statistically significant ($p = 0.429$).

<i>Change in Fear/Insecurity</i>	Increase	Decrease	No change
Suprelorin Implant Group	26.1%	56.5%	17.4%
Surigcal castration Group	27.8%	44.4%	27.8%

Table 18: Changes in Fear/Insecurity during Behavioral Test

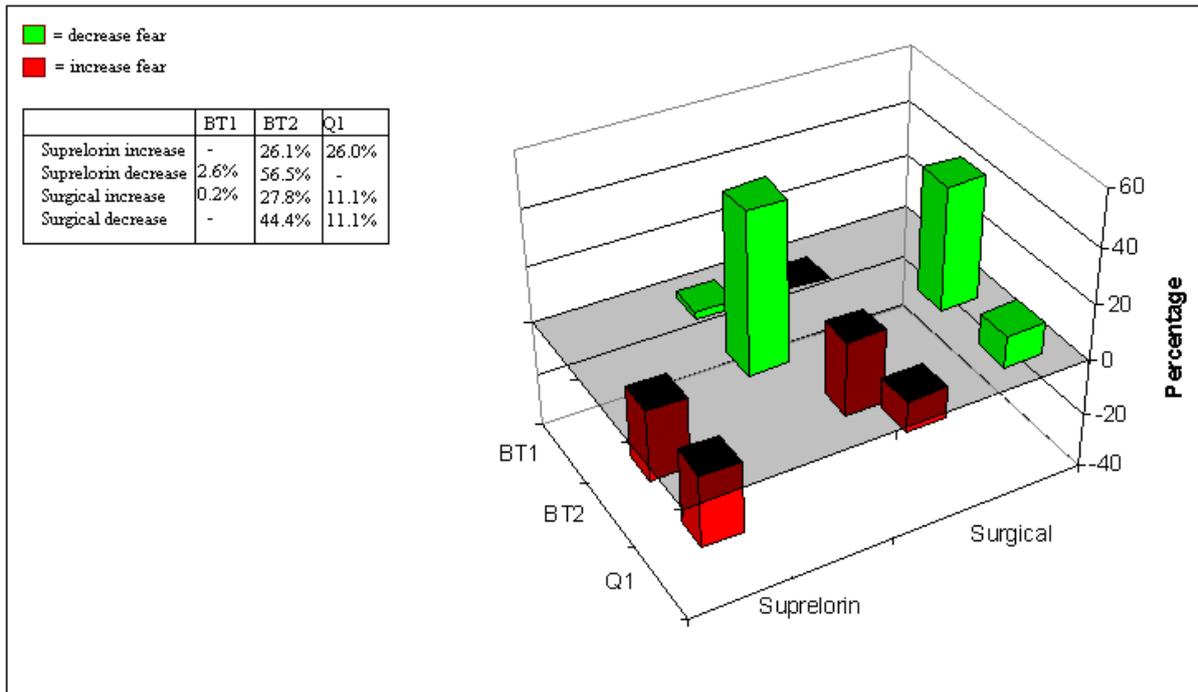


Figure 4: Changes in Fear Responses/Insecurity

BT1: Behavioral Test Entire group. Difference between percentage of fear responses during T=0 and T=2.

BT2: Individual Results of behavioral test. Percentage of dogs that show an increase in fear responses/insecurity and percentage of dogs that show a decrease in fear responses/insecurity.

Q1: Responses to perceptive question. Percentage of owners that noticed an increase in fear responses/insecurity and percentage of owners that noticed a decrease.

Discussion

Aggression

When comparing the results of the behavior test between groups at T=0, we find that in the surgical castration group, significantly more cases of aggression were seen compared to the Suprelorin implant group. This can be explained by the fact that this study was designed as a non-randomized study, resulting in two groups that may not correspond in all fields. If we look at the differences between the compositions of the two groups, one difference that stands out is the representation of the different breeds. In the surgical castration group, the majority of the group consisted of small breeds (shih tzu, maltese, miniature schnauzer, beagle, small mix breeds), while in the Suprelorin implant group most dogs were of larger breeds (retrievers, Rottweiler, bouvier des Flandres). Small dogs are often considered to be more aggressive than large dogs (Arhant et al. 2010, 131-142) so the difference in composition concerning breeds in both groups may explain the significant difference in aggressive behavior between groups at T=0.

Factors such as differences in training of the dogs, socialization of the dogs and consistency of the owners may also play a role in the differences between groups. These factors have however not been assessed, so we are not able to make a valid statement about the contribution of these factors to the differences between the test groups.

Moreover, the biological relevance of the difference between groups at T=0 can be disputed since the overall means concerning aggressive behavior for the suprelorin implant group and the surgical castration group were similar (3.99 and 3.95 respectively, 4=not aggressive). So even though there was a significant difference between the number of cases in which aggression was seen, it is fair to say that overall both test groups showed little to no aggression during the behavior test at T=0.

When comparing the results of the behavior tests at T=0 and T=2 within groups, we find no significant changes in the Suprelorin implant group nor in the surgical castration group.

Also, no significant differences were found between groups concerning the answers given to the perceptive question in the questionnaire at T=2. This shows that even though a significant difference was found between groups at T=0, the changes that occurred following castration in both groups are similar.

In the Suprelorin implant group, 43.5% noticed a decrease of aggression towards other male dogs, in the surgical castration group 47.0% noticed a decrease. These percentages are somewhat lower than findings in previous studies in which decreases of inter male aggression range from 50% – 75% (Hopkins, Schubert, and Hart 1980, 1108-1110; Maarschalkerweerd et al. 1997, 617-619; Beaver 1983, 35-43; Hart 1979, 461-465). However, the interval between the treatment and the interview in some of these previous studies was longer compared to the interval in our study (Hopkins: 27 months, Maarschalkerweerd: 6-12 months). Considering our shorter interval, it might be possible that some of the changes in our dogs haven't occurred yet at the time the questionnaire was filled out.

In the study performed by Hopkins et al. the changes were divided into a rapid or a gradual decline. A gradual decline was considered a decline which was seen after approximately 6 months. That means that in our case gradual declines have not yet occurred and we should compare our

results to the percentage of rapid declines mentioned in the study. If we do so, we find that our results correspond to the results of Hopkins et al, since 38% of the dogs in their study showed a rapid decline of inter-male aggression (Hopkins, Schubert, and Hart 1980, 1108-1110).

When we compare the answers of the perceptive question to the changes that were observed during the behavior test, we can see that many owners reported a decrease of aggression, while the percentages of dogs that showed a decrease of aggression during the behavioral test were much smaller. This can be explained by the fact that the perceptive question was aimed specifically towards inter-male aggression, while in the behavioral test no distinction was made between the different types of aggression. During the behavior test we assessed aggressive behavior in general and did not perform a test which was aimed specifically at inter male aggression. Although such a test may have lead to interesting results, we intentionally chose not to add such a test to our study, since we did not want to jeopardize the safety of the dogs, their owners and the researchers which were involved. It is very likely that the aggressive behavior which was seen during the behavior test was not inter-male aggression but a different type of aggression, fear-induced aggression for instance. This suggestion supported by the fact that in many cases, the aggressive behaviors witnessed during the behavior test were accompanied by a fear response.

Sexual Behavior

Several studies have shown that surgical castration as well as chemical castration leads to a decrease in sexual behavior (Maarschalkerweerd et al. 1997, 617-619; Goericke-Pesch et al. 2010, 920-926). However, in our study we did not find a significant decrease in sexual behavior in the Suprelorin implant group nor in the surgical castration group when assessing the results of the behavioral test.

This might be due to the design of the test- situations. The stimuli that were used to provoke sexual behavior might not have been efficient enough. It is imaginable that during test 4, the scent of the cloth might have been influenced by weather conditions such as the direction of the wind, presence of snow, presence of rain, etcetera.

Also, a validation test which was performed to assess whether a mock dog is fit to impersonate an estrus bitch, showed that mock-dogs are not a suitable alternative for real dogs in order to test sexual behavior of male dogs (Gudde, unpublished results). It can therefore be doubted whether or not the situations used during the behavior test were suitable to assess the effects of castration concerning sexual behavior.

Another possibility might be that the interval between T=0 and T=2 was not sufficient enough for the changes in sexual behavior to become noticeable. Literature has shown that although castration often leads to a decrease of sexual behavior, the time of onset of this change is highly variable (Hart 1974, 383-400). However, if this explanation is correct, we would expect that the absence of a decline of sexual behavior was present during the behavior test as well as in the results of the questionnaire. This is however not the case as a large percentage of the dog owners did notice a decrease.

When looking at question 25 of the questionnaire the number of dogs showing sexual behavior decreased significantly in both the Suprelorin implant group and the surgical castration group. These findings correspond to the available literature, in which decreases of sexual behavior were seen following surgical castration (Maarschalkerweerd et al. 1997, 617-619) and chemical castration (Goericke-Pesch et al. 2010, 920-926).

If we look at the answers to the perceptive questions, we find that significantly more owners from the surgical castration group noticed a decrease in their dog's sexual behavior towards estrus bitches compared to the owners of the Suprelorin implant group (76.5% and 38.1% respectively). Even after GnRH stimulation in the dogs from our study, plasma testosterone concentrations remain low in the surgical castration group (figure 5). The testosterone levels in the Suprelorin implant group show a large amount of variation, although they are not significantly higher. Despite the fact that these levels are highly significantly lower than the pre-treatment levels, these small amounts of testosterone may be responsible for the lesser decrease of sexual behavior towards estrus bitches in some of the dogs in the implant group. We can, however, not be sure of this, since the threshold level of testosterone needed to induce sexual behavior is not known.

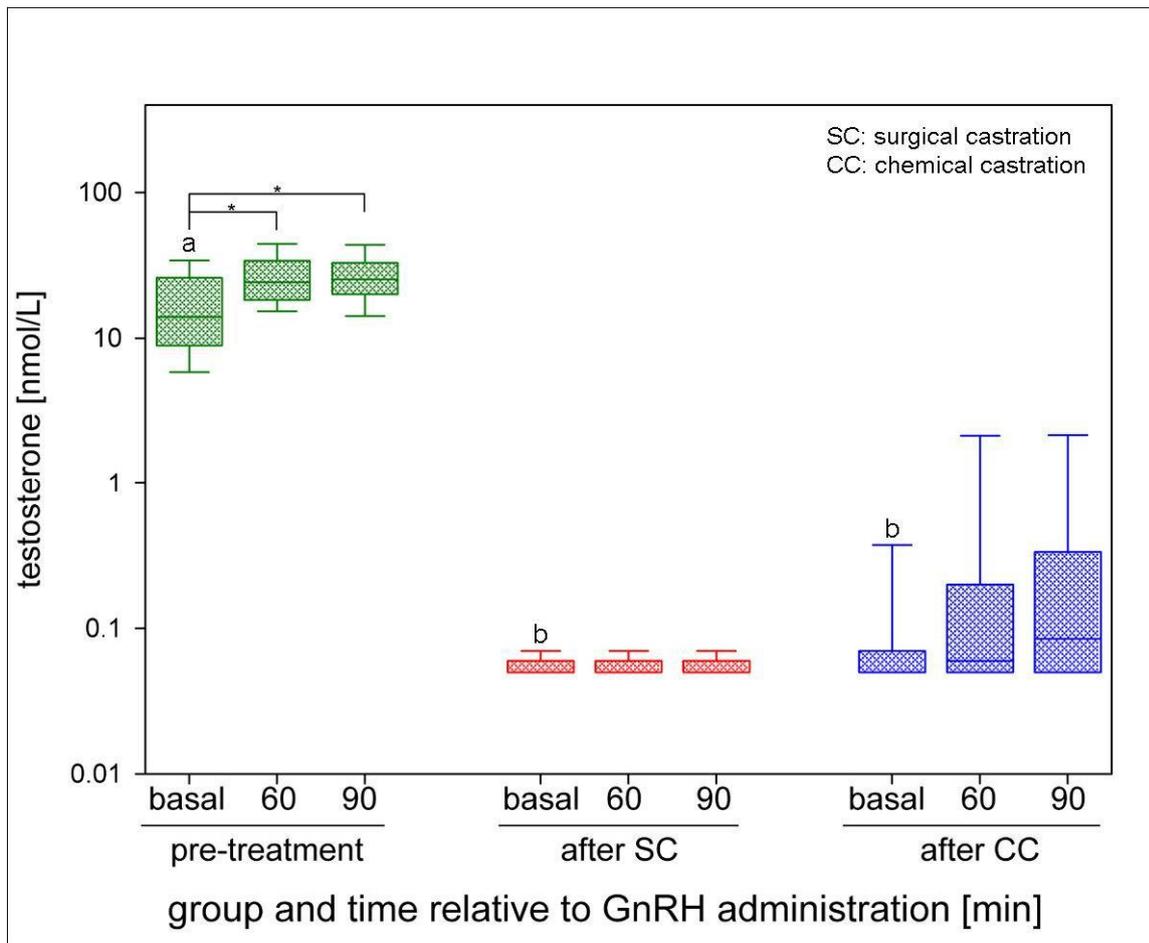


Figure 5: Testosterone response after GnRH Stimulation in the surgical castration group and the suprelorin implant group (de Gier et al., unpublished results)

Play Behavior

Both the Suprelorin implant group and the surgical castration group showed a similar and significant increase of play behavior during the behavior test after treatment. Also, a large percentage of the owners noticed an increase of play behavior in their dogs after castration. These findings do not correspond to the literature available on rats, in which no change in play behavior was seen following post-weaning castration (Smith et al. 1996, 215-226).

It does however correspond to the literature available on ferrets, in which ferrets from the placebo group were found to be less playful than surgically or chemically castrated ferrets (Vinke et al. 2008, 104-121). In this study, the increase in play behavior was explained by the fact that less testosterone leads to less dominant behavior, hence less agonistic interactions, which subsequently leads to more play behavior due to less stressful (competitive) conditions (Vinke et al. 2008, 104-121). Dominance is a sexually dimorphic behavior, male dogs show more dominance than female dogs (Hart and Eckstein 1997, 331-344). This means, that it is likely for this behavior to be altered in dogs following castration. If this is the case, then the above explanation for the increase of play behavior is likely to be true for dogs as well as ferrets. However, in order to be sure of this we would first have to assess our data concerning dominance/submissiveness from our behavior tests and questionnaires.

Fear/Insecurity

In our behavior test, we found no significant differences between the amount of fear responses prior to and after castration. This is true for the surgical castration group as well as the Suprelorin implant group. These findings correspond to a study in dogs, in which no relationship between castration status and fear was found (Döring et al. 2009, 38-43), but do not correspond to the effects of castration in rats and sheep. In these species, studies have shown that castration leads to an increase of fear responses (King, De Oliveira, and Patel 2005, 333-340; Vandenheede and Bouissou 1996, 211-224)

No significant differences could be found in the behavior test between the Suprelorin implant group and the surgical castration group at T=0 and T=2.

We did however find a significant difference between the results of the behavior test and the answers given to the perceptive question in the questionnaire. In the Suprelorin implant group, 56.5% of the dogs showed a decrease of fear responses in the test, while none of the owners noticed a decrease. A significant difference was also found in the surgical castration group, 44.4% of the dogs showed a decrease of fear during the behavioral test, while only 11.1% of the owners noticed a decrease.

These differences can be explained in a couple of ways. During the behavior test we found a decrease in the frequency of the fear responses in a number of dogs. Since the behavior test was filmed and scored at a later time, each situation could be thoroughly observed. As a result of this, all changes were detected, even if they were very small. A small change like this might not have been detected by an owner. This might explain the fact that a significantly smaller proportion of the owners noticed a decrease.

Another possibility is that the situations to which the dogs were exposed during the behavioral test, were not representative of the situations which provoke fear in these dogs. This may explain why the dogs showed little or no fear during the behavioral test, while the owners did consider their dogs to be fearful or insecure.

Finally, it may be possible that during T=2 the dogs were overall less nervous, since they had already been exposed to this situation before. They may have become accustomed to the situation and therefore show less fear. This however, does not seem likely since the time-interval between T=0 and T=2 was quite long (approximately 5 months).

Conclusion

In this study we assessed the effects of surgical and chemical castration on aggression, sexual behavior, play behavior and fear/insecurity in order to determine if the behavioral effects of surgical and chemical castration are comparable.

Fear responses were not influenced by surgical or chemical castration. Play behavior was influenced by surgical as well as chemical castration. In both groups, we saw a significant increase of play behavior during the behavior test. This was supported by the observation of many owners who noticed an increase in play behavior in both groups.

No significant effect was seen concerning aggression during the behavior test. However, the owners of the dogs in both groups did notice a decrease of inter-male aggression. The decreases mentioned by the owners were similar in both groups, but lower than the percentages mentioned in previous studies.

Sexual behavior did not decline significantly during the behavior test in either of the groups. However, many owners did notice a decrease concerning sexual behavior. The effects of chemical castration and surgical castration on sexual behavior towards estrus bitches are however not comparable since significantly more owners from the surgical castration group noticed a decrease in this objectionable behavior.

Overall, this study shows that surgical castration as well as chemical castration does not have a significant effect on aggressive behavior in general or on fear/insecurity and with regard to these behaviors the effects of both treatments are comparable. However, inter male aggression does decrease in many dogs, as was reported by the owners. Also, both treatments cause a similar increase of play behavior in dogs. According to owner reports, sexual behavior towards estrus bitches decreases in both groups, but the number of dogs that show a decrease is significantly greater in the surgical castration group.

Although this study provides some interesting information about the behavioral effects of surgical and chemical castration, further research is needed in order to make a valid statement about the comparability of both treatments. In order to do so, the data which were collected during this study have to be further assessed. These data will provide lots of information about other behavioral elements and will help us to further determine whether or not both treatments are comparable.

Ideally, a second study should be performed during which male dogs are first chemically castrated and will be subsequently surgically castrated. This way, influences such as environment, genetic factors and owner consistency will be eliminated. However, inclusion of a sufficient number of dogs in a study like this will be very hard since you would have to find enough owners who are willing to commit to both treatments, even if chemical castration does not result in behavioral changes that are to the liking of the owners. An alternative would be to perform a study as such in a laboratory setting. However, when you use laboratory dogs, it may be doubtful whether or not the results are also applicable to the dog population in general.

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Appendices

Appendix 1: Questionnaire T=2

Beste deelnemer aan de Deslorelin studie,

Als eigenaar bent u degene die uw hond het beste kent. Ik zou u daarom willen vragen deze vragenlijst zo volledig mogelijk in te vullen. Alle vragen hebben betrekking op de hond die aan het onderzoek meedoet, tenzij er gevraagd wordt om informatie over eventueel aanwezige andere honden in uw huishouden. Het invullen van deze vragenlijst neemt ongeveer 20 minuten in beslag. Een complete vragenlijst zal inzicht geven in de persoonlijkheid van de hond, in dit geval na behandeling. Dit is nodig voor het evalueren van de gevolgen van castratie (chemisch of chirurgisch) bij uw reu.

Zou u bij het beantwoorden van de vragen de situatie willen weergeven zoals deze nu is, dus na de ingreep?

Alvast bedankt!

1. Is de samenstelling van uw gezin veranderd sinds de behandeling?

0 kind het huis uit 0 baby geboren 0 anders, nl

2. Zijn er veranderingen in het aantal honden dat uw huishouden telt sinds de behandeling:* Ja / Nee

Zo ja:

A. Hoeveel:.....

B. Vul s.v.p. de gegevens in van de honden die nieuw zijn of geen deel meer uitmaken van uw huishouden sinds de behandeling.

Ras Leeftijd in jaren Geslacht Mutatie:

1 REU / TEEF * BIJ / AF*

2 REU / TEEF * BIJ / AF*

3 REU / TEEF * BIJ / AF*

4 REU / TEEF * BIJ / AF*

5 REU / TEEF * BIJ / AF*

6 REU / TEEF * BIJ / AF*

* Doorhalen wat niet van toepassing is.

C. Is het gedrag van uw hond naar uw andere honden veranderd?

0 Nee 0 Ja, namelijk.....

3. Is uw woonomgeving veranderd?

0 Nee 0 Ja, namelijk.....

4. Hoe frequent komt uw hond in contact met kinderen?

5. Hoe frequent komt uw hond in contact met andere honden?

.....

6. A. Hoeveel uur per werkdag is uw hond gemiddeld alleen thuis?.....uur

B. Kan de hond goed tegen alleen zijn? * Ja / Nee/ Onbekend

7. A. Hoe draagt uw hond zijn staart normaliter? * Neutraal /hoog/laag

B. Hoe draagt uw hond zijn oren normaliter? * Plat op de kop/omhoog

(Voor meer informatie over interpretatie van lichaamstaal van uw hond kunt u de bijlage lezen aan het einde van deze vragenlijst)

8. Is de hoeveelheid tijd die u aan uw hond besteedt veranderd sinds de behandeling? * Nee / Ja, nl.....

9. In deze vraag wordt een aantal situaties geschetst. U moet steeds aangeven hoe uw reu op een bepaald persoon of dier reageert. Vul in wat het eerste in u opkomt. Meerdere antwoorden zijn mogelijk.

A. Reactie bij benadering door de baas (persoon waar de reu het beste naar luistert) **::

0 Sociaal / vriendelijk	0 Onderdanig
0 Speels	0 Dominant
0 Angstig / onzeker	0 Niet bekend / nvt
0 Agressief	0 Anders, nl.:

B. Reactie bij benadering door andere volwassen gezinsleden / bekenden **::

0 Sociaal / vriendelijk	0 Onderdanig
0 Speels	0 Dominant
0 Angstig / onzeker	0 Niet bekend / nvt
0 Agressief	0 Anders, nl.:

C. Reactie bij benadering door kinderen in het gezin / bekende kinderen **::

0 Sociaal / vriendelijk	0 Onderdanig
0 Speels	0 Dominant
0 Angstig / onzeker	0 Niet bekend / nvt
0 Agressief	0 Anders, nl.:

D. Reactie bij benadering door vreemde man **::

0 Sociaal / vriendelijk	0 Onderdanig
0 Speels	0 Dominant
0 Angstig / onzeker	0 Niet bekend / nvt
0 Agressief	0 Anders, nl.:

E. Reactie bij benadering door vreemde vrouw **::

0 Sociaal / vriendelijk	0 Onderdanig
0 Speels	0 Dominant
0 Angstig / onzeker	0 Niet bekend / nvt
0 Agressief	0 Anders, nl.:

F. Reactie bij benadering door vreemd kind **::

0 Sociaal / vriendelijk	0 Onderdanig
0 Speels	0 Dominant
0 Angstig / onzeker	0 Niet bekend / nvt
0 Agressief	0 Anders, nl.:

G. Reactie bij benadering door vreemde niet gecastreerde reu **::

0 Sociaal / vriendelijk	0 Onderdanig
0 Speels	0 Dominant
0 Angstig / onzeker	0 Niet bekend / nvt
0 Agressief	0 Anders, nl.:

H. Reactie bij benadering door vreemde gecastreerde reu **::

0 Sociaal / vriendelijk	0 Onderdanig
0 Speels	0 Dominant
0 Angstig / onzeker	0 Niet bekend / nvt
0 Agressief	0 Anders, nl.:

I. Reactie bij benadering door vreemde niet loopse teef **::

0 Sociaal / vriendelijk	0 Onderdanig
0 Speels	0 Dominant
0 Angstig / onzeker	0 Niet bekend / nvt
0 Agressief	0 Anders, nl.:

J. Reactie bij benadering door vreemde loopse teef **:

- | | |
|--|---|
| <input type="checkbox"/> Sociaal / vriendelijk | <input type="checkbox"/> Onderdanig |
| <input type="checkbox"/> Speels | <input type="checkbox"/> Dominant |
| <input type="checkbox"/> Angstig / onzeker | <input type="checkbox"/> Niet bekend / nvt |
| <input type="checkbox"/> Agressief | <input type="checkbox"/> Anders, nl.: |

10. In welke mate vertoont uw hond agressie als zijn voerbak/kluif afgepakt wordt? **

- De hond vertoont geen agressieve gedragingen.
- De hond valt uit en bijt.
- De hond blaft, fixeert (= strak aankijken) en blijft aanstaren.
- De hond gromt.
- De hond toont zijn tanden en hapt af en toe.
- Niet bekend / nvt

11. A. Welke van de onderstaande dingen / situaties vindt uw hond beangstigend? Meerdere antwoorden zijn mogelijk **.

- | | |
|---|---|
| <input type="checkbox"/> Harde geluiden | <input type="checkbox"/> Onverwachte bewegingen |
| <input type="checkbox"/> Onweer | <input type="checkbox"/> Vliegenmepper |
| <input type="checkbox"/> Paraplu | <input type="checkbox"/> Bij andere mensen thuis |
| <input type="checkbox"/> Stofzuiger | <input type="checkbox"/> Contact met jonge kinderen |
| <input type="checkbox"/> Vuurwerk | <input type="checkbox"/> Contact met andere dieren |
| <input type="checkbox"/> Winkelcentrum | <input type="checkbox"/> Contact met andere honden |
| <input type="checkbox"/> Drukke autostraat | <input type="checkbox"/> Op een hondenspeelveldje |
| <input type="checkbox"/> In de auto | |
| <input type="checkbox"/> In de bus / trein / ander openbaar vervoer | |
| <input type="checkbox"/> Anders, nl.: | |

B. Hoe gedraagt de reu zich dan in die situatie waarin dit angstgedrag het duidelijkst gezien wordt? **

- Mijn reu vertoont de hier volgende gedragingen: wijken, vluchten, wegstijven, kop laag houden, staart laag dragen (soms tussen de poten), mondhoeken ver naar achteren, oren in de nek leggen en piepen of janken
- Zeer duidelijk, verlaagt houding (staart tussen poten), evt. "bevrozen" (verstijven van angst) of vlucht over lange afstand
 - Redelijk duidelijk, verlaagt houding, vlucht over korte afstand
 - Weinig, lage houding, wegstijven van de situatie, geen vlucht
 - Niet, eventueel wel alert (oren neutraal, staart neutraal)

C. Hoe snel herstelt uw hond zich als hij geschrokken is van iets in de omgeving? **

- is minutenlang van slag
- Herstelt vrij langzaam, maar doorgaans wel binnen enkele minuten
- Herstelt snel, binnen 10 seconden
- Herstelt direct, binnen 2 sec.

12. Hoe beweeglijk is uw hond? **

- De hond ligt bijna de hele dag stil.
- De hond ligt meer dan de helft van de dag stil.
- De hond heeft een regelmatige afwisseling tussen rust en activiteit.
- De hond is vaak actief bezig.
- De hond is bijna voortdurend in beweging.
- Niet bekend / nvt

13. A. Speelt uw reu weleens? **

- Nee
- Ja (svp ook B t/m G beantwoorden)

B. Met wie / wat speelt uw reu? ** (meerdere antwoorden mogelijk)

- Een speeltje of een voorwerp (bijv. een handdoek)
- Iemand binnen het gezin
- Een ander persoon
- Een hond
- Een ander dier
- Anders, nl.:

C. Wie begint er met spelen? **

- De hond zelf
- Iemand binnen het gezin
- Een ander persoon
- Een andere hond of een ander dier
- Wisselend

D. Hoe speels is uw reu? **

(Voor meer informatie over interpretatie van lichaamstaal van uw hond, o.a. speelsheid, kunt u de bijlage lezen aan het einde van deze vragenlijst)

- Hij speelt (vrijwel) nooit
- Hij is weleens speels, als een ander het spel uitdaagt, maar dan niet constant en ook niet erg intensief (komen, kwispelen, speels happen etc.)
- Hij is weleens speels, daagt ook zelf uit tot spelen, maar doet dat niet constant en niet erg intensief (komen, kwispelen, speels happen etc.)
- Hij is zeer speels, daagt vaak zelf uit tot spelen met hoge intensiteit (komen, kwispelen, speels happen etc.)

E. Is de hond meer of minder fanatiek geworden met spelen ten opzichte van de periode dat hij een pup was? **

- nvt, niet als pup gehad
- Nee, geen verandering
- Ja, de hond is minder fanatiek geworden, op een schaal van 1 tot 4:
Een beetje minder fanatiek 0 0 0 0 Veel minder fanatiek
- Ja, de hond is juist fanatieker geworden, op een schaal van 1 tot 4:
Een beetje fanatieker 0 0 0 0 Veel fanatieker

F. Vertoont uw hond de volgende gedragingen bij het spelen? **

- Poot opleggen.
- Gromgeluiden maken.
- Vastpakken van / zachtjes bijten in bijv. de handen.
- Prooi schudden
- Prooi verscheuren
- Nee, de hond vertoont geen van deze gedragingen.

G. Wat doet uw hond als u bij het spelen op de grond gaat liggen? **

- Hij loopt weg
- Hij gaat spel uitdagen
- Hij probeert onder de baas te kruipen en piept en jankt eventueel
- Hij springt op de baas af en gaat er overheen hangen (staart redelijk hoog gedragen)
- Anders, nl.:

14. Ligt uw reu regelmatig op de bank? * Ja/ Nee

15. Ligt uw reu regelmatig op uw voeten? * Ja / Nee

16. Waar slaapt uw hond 's nachts? **

- In de schuur/garage/kennel of andere ruimte buiten het woonhuis, nl:.....
- In de woonkamer
- In de slaapkamer bij één of meerdere gezinsleden
- Anders, nl

17. **A. Hoeveel slaapt uw hond overdag? ***uur / Onbekend

B. Hoeveel slaapt uw hond 's nachts?* Hele nacht/.....uur/ Onbekend

18. **A. Hoe vaak wordt de hond uitgelaten?.....**keer/ per dag

B. Hoe lang per keer(gemiddeld)?.....min./keer

C. Aangelijnd? * Ja, vrijwel altijd / wisselend / Nee, bijna nooit

19. **Wanneer eet uw hond over het algemeen? ***

Nadat u zelf hebt gegeten / voordat u zelf hebt gegeten / tegelijk/ anders,

nl.:.....

20. **Bijt de hond u wel eens in uw handen bij het spelen of bij een begroeting? ***

Ja / Nee

21. **Trekt de hond aan de lijn bij het wandelen? ****

0 Ja, de lijn staat constant strak gespannen; hond hangt in lijn, ik word meegesleept

0 Ja, de lijn staat regelmatig strak; soms hangt mijn hond in de lijn

0 De lijn staat soms wat strak, maar meestal niet

0 Nee, de lijn hangt continu slap

0 Nee, want mijn hond loopt nooit aan de lijn

22. **A. Hoe voert uw hond het commando *lig of af uit*? ****

0 Niet, want hij kent het commando niet

0 Hij kent het commando wel, maar voert het niet of maar half uit

0 Hij voert het commando uit na aandringen met fysiek contact

0 Hij voert het commando uit na aandringen met de stem

0 Hij voert het commando direct uit

B. Hebt u met uw hond getraind op gehoorzaamheid sinds de behandeling? * Ja / Nee

Indien ja, s.v.p. ook C en D beantwoorden.

C. Hoe hebt u getraind op gehoorzaamheid? (meerdere opties zijn mogelijk) **:

0 Gehoorzaamheidscursus (volwassen hond)

0 Individuele begeleiding door gedragsdeskundige

0 Zelfstandig

0 Anders, nl.:

D. Bent u met uw hond op gedragstraining gegaan?

Ja/nee

Cursus: _____

23.

Is uw hond waakzaam? * Ja / Nee

Reageert hij op de postbode? * Ja / Nee

Reageert hij vanuit huis op voorbijgangers? * Ja / Nee

24. **Vindt u uw hond een aandachtvrager? * Ja/ Nee**

25. **A. Toont uw reu weleens seksueel gedrag? * Ja/Nee**

Onder seksueel gedrag verstaan we: bestijgen van en/of rijden op mensen, dieren of voorwerpen, waarbij ook de penis uitgeschacht kan worden en waarbij eventueel sprake kan zijn van ejaculeren.

Indien ja, s.v.p. ook B en C beantwoorden.

B. Waarop is het seksuele gedrag van uw reu gericht? ** (meerdere antwoorden zijn mogelijk)

- Voorwerpen (bijvoorbeeld speelgoed, kussens etc.)
- Teven die niet loops zijn
- Teven die wel loops zijn
- Gecastreerde reuen
- Intacte (niet gecastreerde) reuen
- Kinderen
- Volwassen vrouwen
- Volwassen mannen
- Anders, nl:.....

C. Met welke frequentie vertoont uw reu duidelijk seksueel gedrag?

- meerdere keren per week, spontaan, zonder aanwijsbare stimulus (bijv. een loopse teef)
- meerdere keren per maand, spontaan; met een duidelijke toename als er een stimulus is (bijv. loopse teef in de buurt)
- af en toe spontaan; met een duidelijke toename als er een stimulus is (bijv. loopse teef in de buurt)
- zelden of nooit spontaan, alleen bij duidelijke stimulus (bijv. loopse teef)

D. Is uw hond aantrekkelijk geworden voor andere reuen? * Ja/Nee

26. Bakent uw reu zijn territorium af tijdens het uitlaten? ** (markeren m.b.v urine / vaak poot optillen om te plassen)

- Nooit, mijn reu urineert slechts 1 of enkele keren per wandeling
- Soms, mijn reu tilt zijn poot met name vaak op als er andere reuen in de buurt zijn
- Altijd, mijn reu is altijd gedurende de gehele wandeling bezig met het zetten van geurvlaggen, tilt erg vaak zijn poot op
- Onbekend

27. Loopt uw hond weleens weg? **

- Nooit, mijn hond blijft altijd bij mij in de buurt als hij los loopt en komt als ik hem roep.
- Soms, af en toe loopt mijn hond bij mij weg, maar hij is altijd binnen enkele minuten weer terug.
- Vaak, mijn hond loopt vaak bij mij weg, soms ben ik hem langere tijd (uren/dagen) kwijt.
- Ik laat mijn hond nooit (meer) loslopen, omdat hij vaak wegloopt
- Anders, nl:.....

28. A. Aandacht trekken van de hond tijdens het uitlaten (als hij niet aan de lijn loopt). **

- Mijn hond reageert altijd vlot als ik hem roep wanneer hij buiten losloopt.
- Mijn hond reageert meestal vlot als ik hem roep, maar onder bepaalde omstandigheden niet of nauwelijks (*ga door naar vraag 27 B*)
- Mijn hond reageert zelden vlot als ik hem roep, pas na meerdere herhalingen en stemverheffing lijkt hij mij te horen en reageert hij op de gegeven commando's.
- N.V.T., want mijn hond loopt buiten altijd aangeliend.

B. Indien u bij vraag 28 A de tweede optie hebt aangekruist, graag deze vraag beantwoorden (meerdere antwoorden zijn mogelijk): **

- Mijn hond reageert niet als ik hem roep wanneer hij:
- een loopse teef tegenkomt
- een andere reu tegenkomt

- 0 een andere hond tegenkomt en gaat spelen
 - 0 iets interessant ruikt, hij blijft dan snuffelen / volgt een spoor
 - 0 wild ziet / ruikt
 - 0 iets eetbaars heeft gevonden
 - een speeltje (bal, stok, etc.) heeft dat hij niet wil afgeven
 - 0 anders, nl.....
- (s.v.p. zelf een situatie schetsen waarin uw hond niet komt)*

29. Eet uw hond wel eens van de straat? * Ja / Nee

In vraag 30 t/m 37 wordt gevraagd naar veranderingen sinds de start van behandeling aan het begin van dit onderzoek (castratie of implantatie).

30. In hoeverre vindt u dat agressie van uw hond naar andere reuen is afgenomen? **

- 0 Het gedrag is toegenomen
- 0 Hetzelfde gebleven, geen verandering
- 0 Minder dan 50% afgenomen
- 0 Meer dan 50% afgenomen
- 0 Meer dan 90% afgenomen
- 0 Niet van toepassing, mijn hond was en is nooit agressief naar andere reuen

31. In hoeverre vindt u dat het seksueel gedrag van uw hond naar loopse tevenis afgenomen?

- 0 Het gedrag is toegenomen
- 0 Hetzelfde gebleven, geen verandering
- 0 Minder dan 50% afgenomen
- 0 Meer dan 50% afgenomen
- 0 Meer dan 90% afgenomen
- 0 Niet van toepassing, mijn hond toont en toonde nooit seksueel gedrag richting teven

32. In hoeverre vindt u dat het weglopedrag van uw hond is afgenomen?

- 0 Het gedrag is toegenomen
- 0 Hetzelfde gebleven, geen verandering
- 0 Minder dan 50% afgenomen
- 0 Meer dan 50% afgenomen
- 0 Meer dan 90% afgenomen
- 0 Niet van toepassing, mijn hond liep en loopt nooit weg
- 0 Niet van toepassing, mijn hond loopt nooit los

33. In hoeverre vindt u dat de voorhuidontsteking van uw hond is afgenomen? **

- 0 De voorhuidontsteking is nieuw ontstaan of toegenomen
- 0 Hetzelfde gebleven, geen verandering
- 0 Minder dan 50% afgenomen
- 0 Meer dan 50% afgenomen
- 0 Meer dan 90% afgenomen
- 0 Niet van toepassing, mijn hond had en heeft nooit last van voorhuidontsteking

34. In hoeverre vindt u dat het markeergedrag (urine) van uw hond is afgenomen? **

- 0 Het urinemarkeren is toegenomen
- 0 Hetzelfde gebleven, geen verandering
- 0 Minder dan 50% afgenomen
- 0 Meer dan 50% afgenomen
- 0 Meer dan 90% afgenomen
- 0 Niet van toepassing, mijn hond heeft dat nooit gedaan en doet dat nog steeds niet

35. In hoeverre vindt u dat uw hond angstiger is geworden? **

- 0 Mijn reu is minder angstig geworden
- 0 Hetzelfde gebleven, geen verandering
- 0 Minder dan 50% toegenomen
- 0 Meer dan 50% toegenomen

- Meer dan 90% toegenomen
- Niet van toepassing, mijn hond was en is nooit angstig

36. In hoeverre vindt u dat uw hond beter bereikbaar is? **

- Mijn reu is minder bereikbaar geworden
- Hetzelfde gebleven, geen verandering
- Minder dan 50% toegenomen
- Meer dan 50% toegenomen
- Meer dan 90% toegenomen
- Niet van toepassing, mijn hond was en is nooit onbereikbaar

37. In hoeverre vindt u dat de speelsheid van uw hond is toegenomen? **

- De speelsheid is afgenomen
- Hetzelfde gebleven, geen verandering
- Minder dan 50% toegenomen
- Meer dan 50% toegenomen
- Meer dan 90% toegenomen
- Niet van toepassing, mijn hond was en is nooit speels

Vragen met betrekking tot de fysieke kenmerken van uw hond:

38. Heeft uw reu weleens last van voorhuidontsteking (pussige, geelgroene uitvloeiing uit de voorhuid)? **

- Ja, vaak / altijd
- Ja, soms
- Nee, nooit
- Onbekend

39. Is uw reu incontinent voor urine? **

- Nee
- Ja

40. Wat is uw mening over de eetlust van uw reu? **

- Zijn eetlust is goed
- Zijn eetlust is matig
- Zijn eetlust is slecht

41. Wat is uw mening over de vachtconditie van uw reu? **

- De vachtconditie van mijn hond is goed
- De vachtconditie van mijn hond is matig
- De vachtconditie van mijn hond is slecht

42. Heeft uw reu nadelige gevolgen ondervonden van de behandeling die hierniet zijn vermeld?

Bijzonderheden / opmerkingen ten aanzien van het gedrag van mijn hond:

.....

.....

.....

.....

.....

.....

.....

.....

.....

.....

Opmerkingen ten aanzien van de gezondheid van mijn hond:

.....

.....

.....

.....

.....

Dank voor het invullen van deze vragenlijst!

Appendix 2: Description of behavioral tests.

1. meeting and approaching a stranger (male)

Description: the owner will enter the testing area with the dog on a leash. In the testing area the dog and owner will meet a stranger (man). The man will first greet the owner, after that, he will squat down and call the dog and let the dog sniff his hands. This test will last for two minutes; during the second minute the man will try to stimulate play behavior.

2. Exploration of a new environment

Description: the dog will be let of the leash and will be given the opportunity to explore the test environment. This test will last for three minutes.

3. Walking outside on a leash

Description: At first, the owner will walk the dog without giving any commands. Hereafter, the owner will give the dog the command 'follow' and they will walk the same course once again.

4. Walking outside on a leash with an olfactory stimulus present

Description: This test is almost the same as test 3, with the exception that during this test, there is an attractive olfactory stimulus present somewhere along the course (tampon with discharge of a bitch in heat). This test, again, consists of two phases, phase one: walking the dog without a command, phase two: walking the dog with the command 'follow'.

5. Performing basic commands outside: Sit, Down, Come

Description: The owner will stand in front of the dog and give the command: 'sit'. The dog has to remain seated for twenty seconds. After twenty seconds, the command 'down' will be given. This position also has to be maintained for 20 seconds. The final part of this test is asking the dog to sit down, walking away from the dog (dog has to remain seated) and then calling the dog: 'come'.

6. Walking on a leash (inside)

Description: At first, the owner will walk the dog without giving any commands. Hereafter, the owner will give the dog the command 'follow' and they will walk the same course once again.

7. Walking on a leash with an olfactory stimulus present (inside)

Description: This test is almost the same as test 6, with the exception that during this test, there is an attractive olfactory stimulus present somewhere along the course (tampon with discharge of a bitch in heat). This test, again, consists of two phases, phase one: walking the dog without a command, phase two: walking the dog with the command 'follow'.

8. Performing basic commands inside: Sit, Down, Come

Description: The owner will stand in front of the dog and give the command: 'sit'. The dog has to remain seated for twenty seconds. After twenty seconds, the command 'down' will be given. This position also has to be maintained for 20 seconds. The final part of this test is asking the dog to sit down, walking away from the dog (dog has to remain seated) and then calling the dog: 'come'.

9. Petting the dog's back by the owner

Description: The dog's owner will squat down next to the dog and will stroke the dog from his head to the end of his back during thirty seconds. This test is performed to assess if the dog will accept social dominance. During this test, the dog will be on a leash constantly.

10. Holding the dog down on its back by the owner (restraint)

Description: The owner will squat down next to the dog and gently roll the dog onto its back. The dog will be held in this position for thirty seconds. This test was performed to simulate the enforcement of passive submission.

11. Playing with a floss-rope

Description: During the first phase of this test, we will assess if play behavior can be provoked. After one minute of playing, the owner will tell the dog to let go of the floss-rope (phase 2). This part of the test was designed to assess whether or not the dog will accept dominance.

12. Owner lying down on the floor

Description: The owner will lay down flat on the ground. The dog will be let of the leash and his behavior will be observed for thirty seconds.

13. Meeting an unfamiliar child (doll)

Description: A researcher will enter the testing area with the child-doll (toddler). They will approach the dog and try to let the doll make contact with the dog. The doll will stroke the dogs back for approximately ten seconds. During this test, the dog will be on a leash constantly.

14. Owner staring at the dog

Description: The owner will (try to) look the dog straight in the eye for twenty seconds. During this test, the dog will be secured to a wall and the owner will keep a safe distance from the dog.

15. Stranger staring at the dog

Description: a researcher will (try to) look the dog straight in the eye for twenty seconds. During this test, the dog will be secured to a wall and the researcher will keep a safe distance from the dog.

16. Visual Stimulus (umbrella)

Description: the researcher will walk past the dog twice holding an umbrella. After this, he will open the umbrella and then open and close it quickly a few times, as if he is shaking the rain of the umbrella. During this test, the dog will be on a leash constantly.

17. Acoustic Stimulus (Rattle)

Description: the researcher will walk past the dog twice holding a rattle. After this, he will make noise with the rattle. During this test, the dog will be on a leash constantly.

18. Meeting a bitch in heat (mock-dog)

A mock dog will be placed in the testing area. A tampon with discharge from a bitch in heat is attached to the mock dog's perineum. The owner and dog will enter the room and the dog will be able to interact with the mock-dog for one minute. Hereafter, the owner will give the command: 'Come' and take the dog away from the mock-dog. During this test, the dog will be on a leash constantly.

Appendix 3: Ethogram of Behavioral Test

Approach

Dog does not approach	0
Dog approaches slowly (30s>), stays at a small distance	1
Dog approaches slowly (30s>), physical contact	2
Dog approaches immediately, physical contact	4

Sociability

No social behavior	0
Alert, no physical contact	1
Looking/alert, tail wagging, no physical contact	2
Attempt to make physical contact, tail wagging, reaching a paw	3
Physical contact, jumping up, wagging the tail	4

Play behavior

No or little play behavior	0
Playful, reactive, not constantly or intensively	1
Playful, pro-active, not constantly or intensively	3
Playful, pro-active, constantly with a high intensity	4

Fright: this is the dog's primary reaction

Severe: posture is lowered, flights long distances, freezing	0
Considerable: lowers his posture, flights short distances	1
Fair: low posture, looking away from the situation, no flight	3
No fright	4

Recovery

No recovery	0
Recovery after 10 seconds but within a minute	1
Recovery within 10 seconds	3
Immediate recovery: within 2 seconds	4

Fear/Insecurity

Severe: lowers posture (tail between legs), freezing, long-distance flight	0
Considerable: lowers posture, short-distance flight	1
Fair: low posture, looking away from the situation, no flight	3
No fear/uncertainty	4

Submissiveness/Dominance

Very dominant: head and tail are high, ears are pointed, legs are stretched. Ambivalence: posture and behavior are conflicting	0
Considerably dominant: head and tail are higher than normal	2
Very submissive: tail between the back legs, licking, squatting through the back legs, ears flat, head low	2
Fairly submissive: tail is low, but not between the legs, ears flat, head low	3
Neutral: neutral and relaxed posture	4

Level of Activity

Lots of activity: dog is constantly walking around/ barking/ jumping up walls or doors	0
Some activity: dog walks around once in a while, annoying vocalisations such as whining and howling	2
Dog does not display any of the behaviors mentioned above	4

Aggressiveness

Aimed attempt to bite: shaking	0
Deep growl, fixating, showing teeth, snapping	1
Growling, fixating, showing teeth	2
Fixating	3
No aggression	4

Posture:

High: overstretched extremities, ears forward, tail up	0
Ambivalent	0
Low	1
Half low	2
Half high	2
Neutral	4

Exploration:

The dog does not explore the testing area	0
Dog explores for a short amount of time (<10% of the testing time, <18 seconds)	1
Dog explores for more than half the time (>90 seconds), but eventually sits or lays down.	2
The dog explores for the entire amount of time (3 minutes)	4

Pulling on the leash:

Leash is constantly under tension: dog 'hangs' in the leash, owner is dragged along,	0
Leash is often under tension: some 'hanging' in the leash	1
Dog pulls on the leash once in a while	2
Dog does not pull on the leash	4

Obedience:

The dog does not know the command	0
The dog does not or only partially follows the command	1
The dog follows the command after physical contact	2
The dog follows the command after insisting by the owner (voice)	3
The dog immediately follows the command	4

Reaction to olfactory stimulus: Sexual behavior:

Notices stimulus, pulls owner along, sniffing, teeth chattering, Salivation	0
Notices stimulus, pulls owner along, sniffing	1
Notices stimulus, looks at it, but follows owner anyhow	3
Dog ignores stimulus	4

Reaction to restraint:

Fights heavily, turns around, bites (not playful), growls	0
Fights heavily, turns around	1
Fights, then surrenders	2
Does not fight the restraint	4

Staring:

Stares back the entire time, aggressive elements	0
Stares back the entire time, no aggressive elements	1
Stares back <10 seconds, then looks away	2
Looks away immediately, but keeps trying to make eye contact	3
Looks away immediately and looks away during 20 seconds	4

Sexual Behavior towards estrus bitch:

Sniffs the perineum, teeth chattering, salivation, mounting, attempt to mate	0
sniffs the perineum, teeth chattering, salivation, no mounting	1
sniffs the perineum, then loses interest	3
None of the above behaviors	4