

Surgery Not Required: Current and Future Options in Fertility Control of Dogs and Cats

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Mission: To expedite the successful introduction of methods to non-surgically sterilize dogs and cats and to support the distribution and promotion of these products to humanely control cat and dog populations worldwide.

acc-d.org



Priorities for Non-Surgical Fertility Control

- Safe and effective
- Permanent or long-term
- •Deliverable in a single injection or treatment
- Approaches for dogs and cats / male and female
- Documented effects on behavior and health
- Affordable

ACC&D Resources





February 2013 Product Profile and Position Pape

Esterilsol[™]/Zeuterin[™] is a non-surgical sterilant for male dogs delivered via intratesticular injection. The active ingredient is zinc gluconate neutralized by arginine. The formulation causes permanent infertility in one treatment. It is also known as "zinc neutering".



Why nonsurgical sterilization instead of traditional spay/neuter?











To save more lives....



...we need more options.

Let's explore the options.

- On the horizon
- Available now

How is ACC&D paving the way for these options?



Michelson Prize and Grants Program in Reproductive Biology

Founded in 2009





\$50M available for research grants to develop a non-surgical sterilant for male and female dogs and cats.



"A nonsurgical sterilant could reduce the global population of homeless dogs and cats, but there hasn't been money to develop one—until now."



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18 SEPTEMBER 2009 VOL325 SCENCE www.science.emug.ceg Publication.execution.cemug.ceg

ACC & D Alliance for Contraception in CATS & DOGS

September 2009 issue of Science, "A cure for euthanasia?"

Goal

- Reduction or elimination of deaths of
 healthy shelter cats and dogs in the United
 States
- Successful product will likely be made
 available world wide for feral cat and dog
 control



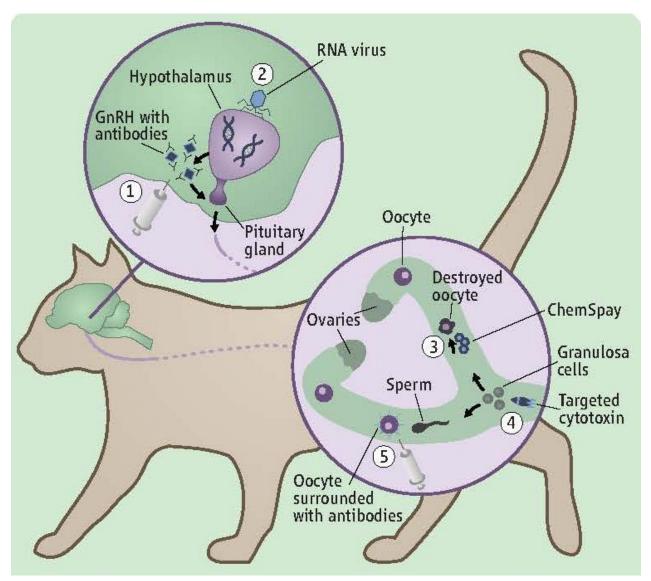


Research Approaches

Immunocontraception

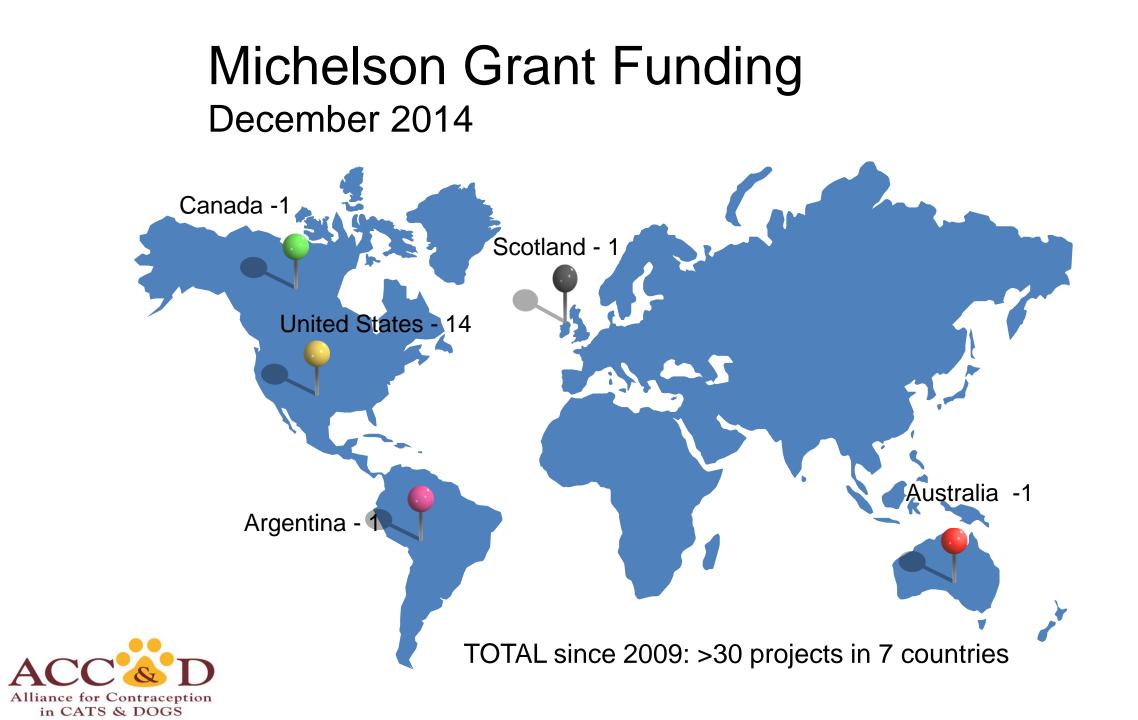
High dose, long-term GnRH agonists Targeted delivery of cytotoxins

Gene silencing/gene therapy





September 2009 issue of Science, "A cure for euthanasia?"



\$25M Prize

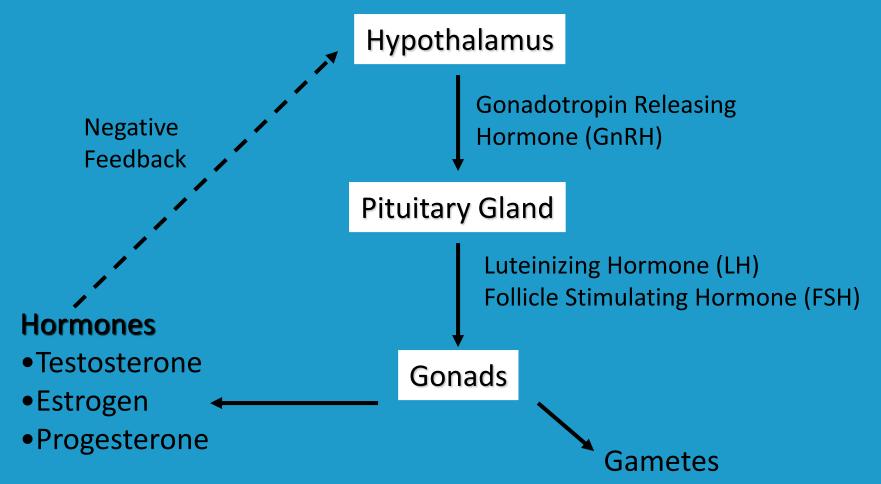
- Single-dose, permanent, nonsurgical sterilant
- Safe and effective in male and female cats and dogs
- Ablates sex steroids and/or their effects
- Suitable for administration in a field setting
- Viable pathway to regulatory approval
- Reasonable manufacturing process and cost



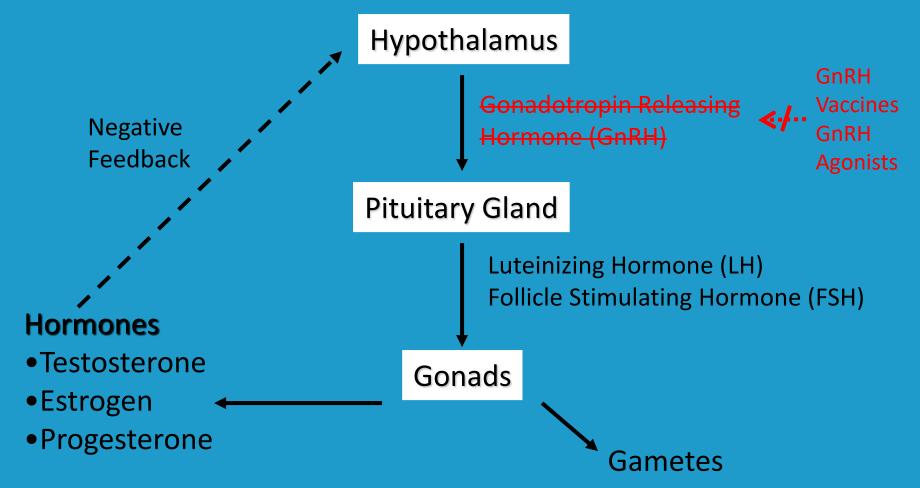




Reproductive Control



Reproductive Control



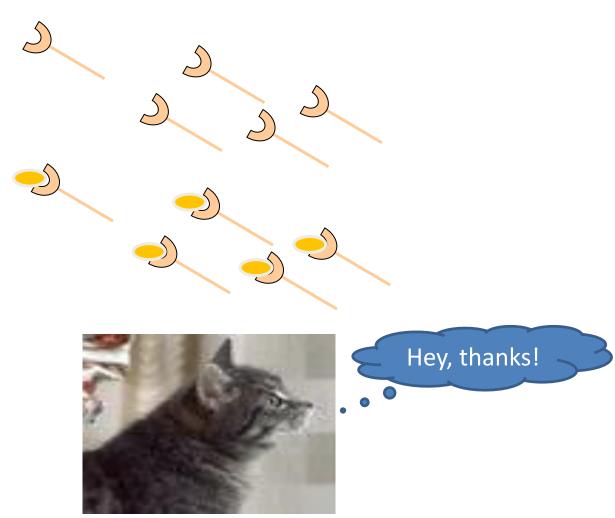
Slimmy receives a rabies vaccine.

Her body makes antibodies against the virus.

If exposed to the virus, circulating antibodies bind it. The virus does not reach its receptor.

Slimmy is not infected.

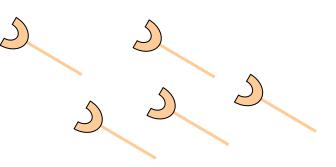






Her body makes antibodies against her own GnRH.





When her hypothalamus releases GnRH, antibodies bind it.

GnRH does not reach its receptors in the pituitary.

Cerebellum Spinal cord Medulla oblongata

FSH and LH are not released.

Slimmy is contracepted.



GonaCon

- Overview:
 - Developed by USDA-NWRC
 - EPA registered for use in white-tailed deer (2009) and wild horses and burros (2013)
 - Contraceptive effect in several other species



GonaCon

Dr. Levy's studies demonstrated safety, efficacy & suppression of sexual behaviors in male & female laboratory cats.



Photo: Julie Levy

Results in Male Cats

- Single dose GonaCon
- 24 adult male cats 2 studies
 - Antibody titer, testosterone and semen analysis
 - Secondary sex characteristics



- Breeding trial mean time to successful breeding was 12 mos. for responders
- 3-year follow-up; no injection site issues

Secondary Male Sex Characteristics



Testicle shrinkage

Loss of penile spines

Slide: Courtesy of Dr. Julie Levy, Maddies' Shelter Medicine Program, University of Florida College of Veterinary Medicine.

Results in Female Cats

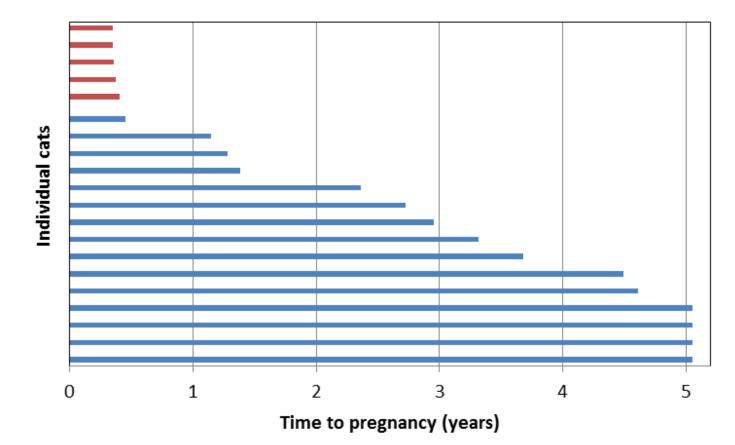
- 20 adult female cats
- Single dose GonaCon
- Monthly measurements
 - GnRH antibody titers
 - Estrogen, progesterone
- Breeding trial
- 5-year follow-up



Photo: Julie Levy

GonaCon treated female cats: Median time to pregnancy 39.7 mos.

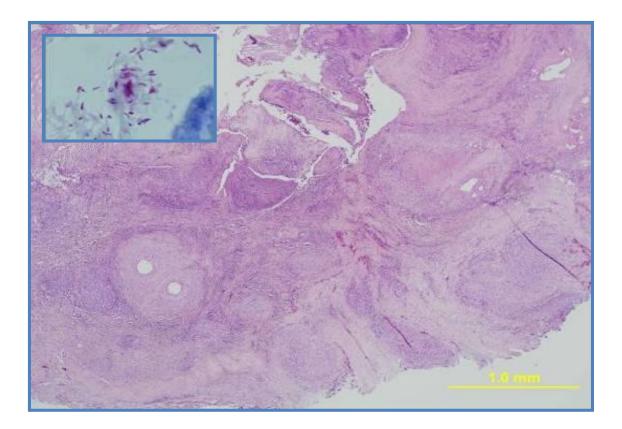
Sham-treated cats
GnRH-vacinated cats



Slide: Courtesy of Dr. Julie Levy, Maddie's Shelter Medicine Program, University of Florida College of Veterinary Medicine

Injection-Site Reactions

- Non-painful granulomatous masses
- Late onset (~24 months) in 6/20 cats



Slide: Courtesy of Dr. Julie Levy, Maddies' Shelter Medicine Program, University of Florida College of Veterinary Medicine.

Why we are excited about GonaCon

- Contraceptive vaccine safe & effective in female cats
 - 3+ year average duration; suppression of sexual behaviors
- Practical for field administration
- Potentially very affordable





ACC&D is working to realize GonaCon's potential for free-roaming cats.

Deslorelin / Suprelorin[®] VIRBAC, formerly Peptech Animal Health

- GnRH agonist
- Delivered by implant
- Approved in Australia, New Zealand and EU for male dogs
- FDA-indexed product to treat adrenal tumors in ferrets



Dogs with No Names

Dr. Judith Samson-French and her team implant dogs on First Nations Reserves with Suprelorin.

Give female dogs a "pregnant pause"

dogswithnonames.com



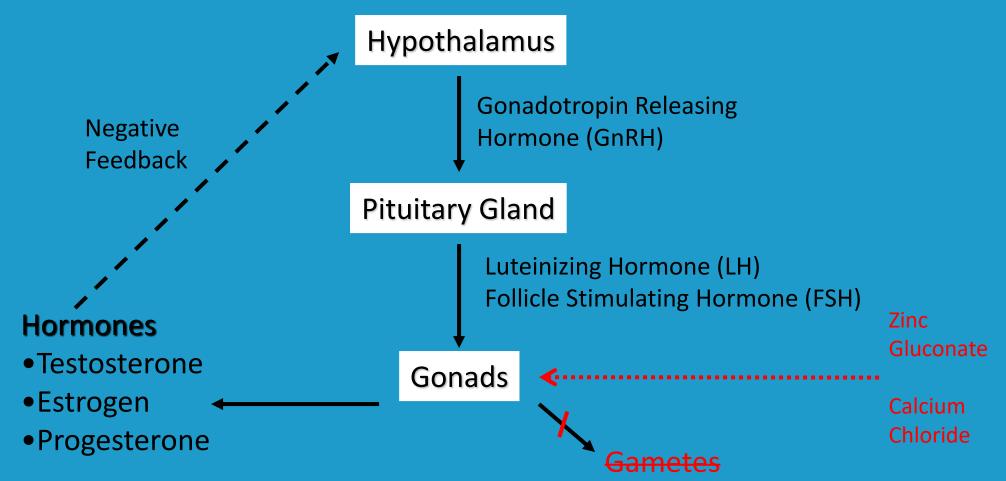
Chemical sterilization

The use of chemical compounds which destroy and/or render ineffective essential component(s) of the reproductive system

Current approaches are for males

 Zinc gluconate neutralized by arginine
 Zeuterin[®]
 Calcium chloride

Reproductive Control



New York Times December 2, 2013

"New Strides in Spaying and Neutering"



Wall Street Journal November 26, 2014

"Too Many Dogs: A Simple Solution"



A cheap, quick, relatively painless procedure could make a big dent in overpopulation. What's stopping it?



A cheap, quick, relatively painless procedure could make a big dent in canine overpopulation. What's stopping it?

3 MONTHS FOR JUST \$1 ACT NOW

http://online.wsj.com/articles/too-many-dogs-a-simple-solution-for-sterilization-1417187544[11/29/2014 10:08:58]

Leoci et al. Acta Veterinaria Scandinavica 2014. 56:62 http://www.actavetscand.com/content/56/1/62



RESEARCH

Open Access

Alcohol diluent provides the optimal formulation for calcium chloride non-surgical sterilization in dogs

Raffaella Leoci^{1*}, Giulio Aiudi¹, Fabio Silvestre¹, Elaine A Lissner² and Giovanni M Lacalandra¹

Abstract

Background: Surgical castration is widely used to sterilize male dogs, but has significant impacts on time to perform the operation, recovery of the animals as well as cost, which can limit population control programs. Previous research has shown intratesticular injection of calcium chloride dihydrate (CaCl-) in saline to be a promising alternative to surgery. However, long-term azoospermia was not maintained at dosages low enough to avoid side effects. In the search for an optimized formulation, the current investigation is the first study on long-term sterilization effects of intratesticular injection of CaCl₂ in either lidocaine solution or alcohol in dogs. CaCl₂ at 20% concentration in lidocaine solution or alcohol was administered via intratesticular injection to proups of 21 dogs each. The treated animals were examined at 2, 6, and 12 months for sperm production, blood levels of testosterone, and side effects; at time zero and 12 months for testicular size and semen volume. The experimentally treated animals were compared to a control group receiving saline injection only.

Results: Testicles of dogs treated with CaCl₂ in either diluent significantly decreased in size. After administration of CaCluin lidocaine solution, sterility was achieved for at least 12 months in 75% of treated doos. However, optimal long-term contraceptive effectiveness was achieved with CaCl₂ in alcohol, which resulted in azoospermia over the 12-month study period. Testosterone levels significantly decreased following treatment with CaCl₂₂ and sexual activity disappeared. Although testosterone returned to baseline levels by 12 months for the group treated with CaCl₂ in lidocaine, dogs injected with CaCl₂ in alcohol had a 63.6% drop in testosterone level, which remained at the low end of physiological range throughout the study. No adverse effects were noted, Conclusions: A single, bilateral intratesticular injection of 20% CaCl₂ in 95% ethanol was a reliable method for induction of sterilization in 18-28 kg male dogs in this study. The approach showed long-term efficacy and reduced sexual behavior. This chemical method of sterilization might provide an effective, efficient alternative to surgical castration that can have positive impacts on dog welfare.

Keywords: Calcium chloride, Canine, Chemical castration, Dog, Nonsurgical sterilization, Population management

Background

governments to reach further with limited resources [1].

An intratesticular injection of calcium chloride dihy Canine overpopulation remains a problem facing many drate (CaCl2) in solution represents a promising method countries throughout the world. Alternative methods to for non-surgical sterilization [2-7]. A previous dosesurgical sterilization that are effective, easy to administer, determination study reported that a 20% solution of CaCl2 safe, and affordable would offer immense benefits, allowing in saline demonstrated good long-term efficacy without animal welfare organizations, public health programs, and the undesirable side effects that occurred with higher dosages [2]. These findings partially confirmed the results of short-term, histology-based studies on CaCl2 by other investigators who used a 20% concentration [3.5-7]. However, when 20% CaCl₂ in saline solution, as typically used for sterilization, was evaluated for efficacy over a longer period, the effect was not permanent: sperm production

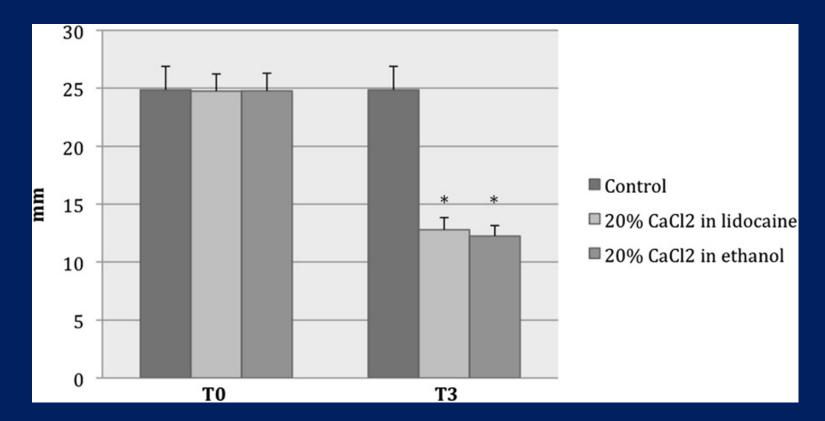
* Correspondence: leocivet@yahoo.it Department of Emergency and Organ Transplantation (DETO), Section of Veterinary Clinic and Animal Production, University of Bari Aldo Moro, SP per Casamassima Icm 3, Valenzano BA, Italy Full list of author information is available at the end of the article.

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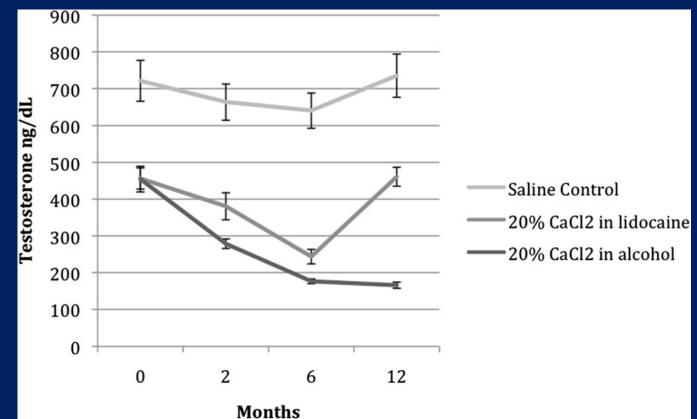
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Results: Testicles of dogs treated with CaCl₂ in either diluent significantly decreased in size. After administration of $CaCl_2$ in lidocaine solution, sterility was achieved for at least 12 months in 75% of treated dogs. However, optimal long-term contraceptive effectiveness was achieved with CaCl₂ in alcohol, which resulted in azoospermia over the 12-month study period.

Testosterone levels significantly decreased following treatment with CaCl₂, and sexual activity disappeared. Although testosterone returned to baseline levels by 12 months for the group treated with CaCl₂ in lidocaine, dogs injected with CaCl₂ in alcohol had a 63.6% drop in testosterone level, which remained at the low end of physiological range throughout the study. No adverse effects were noted.



Changes in testicular width after intratesticular injection of $CaCl_2$. At 12 months (T3) after treatment with $CaCl_2$ (group A and group B), significant reductions in testicular width were observed (*P < 0.001), as compared with no or minimal changes seen in the control (C) group.



Effects of intratesticular injection of $CaCl_2$ on serum testosterone levels over time. Following the injection of $CaCl_2$ in lidocaine solution (group A), testosterone decreased significantly (F = 0.47; P < 0.003) for up to 6 months, although testosterone levels at 12 months returned to baseline. After injection of calcium chloride in alcohol (group B), testosterone levels decreased significantly (F = 65.1, P < 0.001) throughout the 12-month follow-up period.

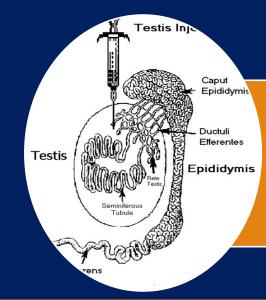
ACC&D Position Statement on Calcium Chloride: • To our knowledge, CaCl₂ has not been reviewed or approved by any regulatory agency for use as an animal sterilant.

 ACC&D believes that the current use of intratesticular CaCl₂ as a sterilant should be considered experimental.

Zeuterin™



Zinc Gluconate Neutralized by Arginine is the only FDA approved non-surgical sterilant for male dogs



A single, virtually painless injection to neuter a male dog, permanently

ZeuterinTM Highlights

Male dogs 3-10 months of age (older pending)

One-time injection into each testicle

Safe & Effective

Permanent and Irreversible

Reduces Testosterone

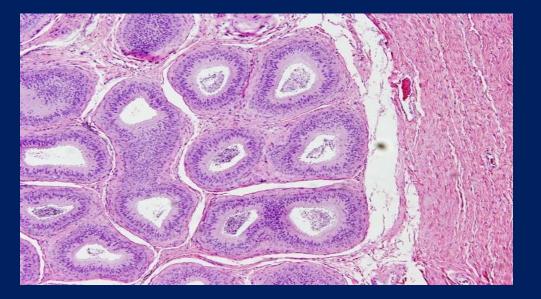
Efficacy

Proven safe and effective

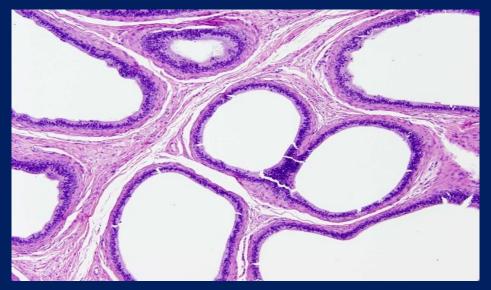
- 99.6% Efficacy
- FDA clinical trial: 223/224 dogs age 3 to 10 months

How do we know Zeuterin is permanent?

Epididymis from untreated dog (100x)



Epididymis from Zeuterin treated dog (100x)

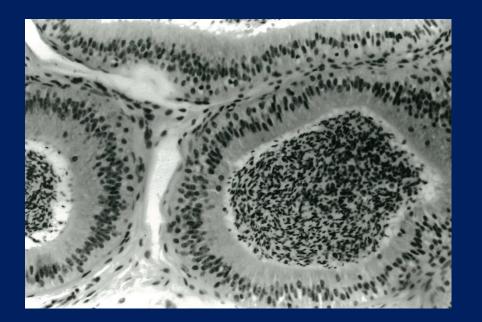


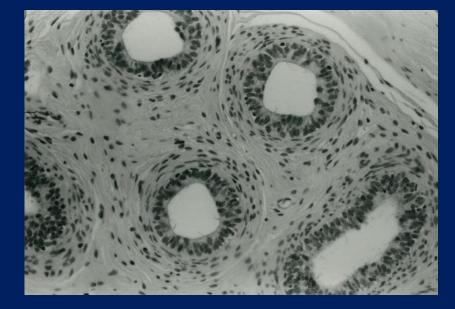
At 30 days post-injection

How do we know Zeuterin is permanent?

Epididymis from untreated dog (100x)

Epididymis from Zeuterin treated dog (100x)

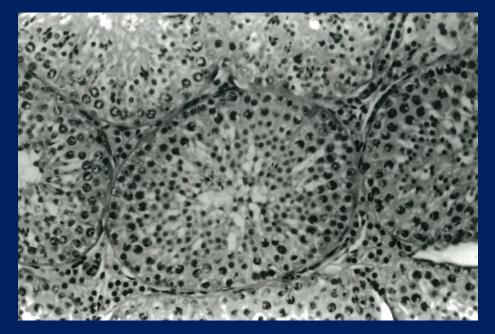




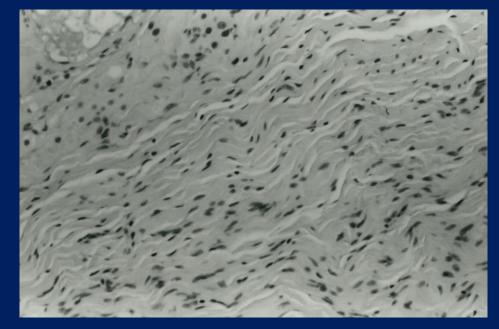
At 24 months post-injection

How do we know Zeuterin is permanent?

Testicle from untreated dog (100x)



Testicle from Zeuterin treated dog (100x)



At 24 months post-injection

Research Behind FDA Approval

- Twelve years of scientific research
- Dosage determination study
- Animal Safety Study
- Clinical Trial 224 dogs 3-10 months old
- Follow-up of dogs in original study

Adverse Reactions Observed During FDA Trial

Local Reactions	No. of Dogs (n = 270)	Percent (%)	Systemic Reactions	No. of Dogs (n = 270)	Percent (%)
Scrotal Pain*	17	6.3%	Neutrophilia	17	6.3%
Scrotal Irritation	3	1.1%	Vomiting**	12	4.4%
Biting and Licking	2	0.7%	Anorexia	11	4.1%
Scrotal Swelling	2	0.7%	Lethargy	6	2.2%
Scrotal Irritation/Dermatitis	2	0.7%	Diarrhea	5	1.9%
Scrotal Ulceration	1	0.4%	Leukocytosis	2	0.7%
Scrotal Infection	1	0.4%			
Dry Scrotal Skin	1	0.4%			
Scrotal Bruising	1	0.4%			
Preputial Swelling	1	0.4%			
Scrotal Sore	1	0.4%	Courtesy Ark Sciences		

* No NSAIDs were used and most scrotal pain was reported on the first two days after injection.

** Vomiting was most commonly seen on the day of the injection, between 1 minute and 4 hours post-injection

Expected Reactions

- Injection technique is critical to prevent adverse reactions
- Normal reactions include:
 - Generally non-painful swelling for 24-48 hours
 - Listlessness for the first 24 hours
 - May or may not vomit during the first 24 hours
- Should return to normalcy within 24 hours

How do we know Zeuterin is painless?

"Pain receptors are considered to be present in the tunicae testis and it would seem reasonable to suppose that, as in other solid organs, the acute pain experienced in testicular injury is due in part to stretching of the capsule."

The Testis, Volume 1, Development, Anatomy, and Physiology. Academic Press, New York and London, 1970; p. 80-81 Chapter 2, Subchapter 5. Hodson, N. Nerves of the Testis, Epididymis, and Scrotum.

Long Term Effects

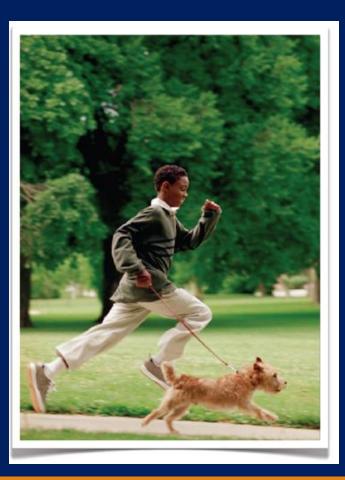
- 30 six-month old dogs treated with Zeuterin were followed for 2 years
- 2 years of routine observations and clinical evaluations
- Permanent reduction of circulating testosterone of 41-52%
- 24 months post injection necropsy concluded all vital organs normal
- Epididymides, testicles and prostate had all reduced in size

Long Term Effects

Zinc Neutering spares the testosterone producing function of Leydig cells, keeping the critical endocrine system intact.

Without spermatogenesis, the testosterone production goes down by 41-52%

Testosterone production remains available for other metabolic functions of the dog



Long Term Effects of Surgical Neutering Show Benefits and Detriments

(Source: Determining the optimal age for gonadectomy of dogs and cats, Margaret V. Root Kustritz, DVM, PhD, DACT)

	Condition	Incidence	Substantial morbidity?	Specific breeds at risk?	
	Benefits				
<	Testicular neoplasms	0.9%	No	No	
<	BPH or prostatitis	75%–80% by 6 years of age	No	No	
	Detriments				
	Complications of surgery	6.1%	No	No	
<	Prostatic neoplasms	0.2%–0.6%	Yes	No	
	ТСС	1.0%	No	Yes*	
	Osteosarcoma	0.2%	Yes	Yes†	
	Hemangiosarcoma	0.2%	Yes	Yes‡	
	CCL rupture	1.8%	Yes	Yes§	
	Obesity	2.8%	No	Yesll	
	Diabetes mellitus	0.5%	No	Yes¶	

* Airedale Terrier, Beagle, Collie, Scottish Terrier, Shetland Sheepdog, West Highland, White Terrier, and Wire Fox Terrier.

† Doberman Pinscher, Great Dane, Irish Setter, Irish Wolfhound, Rottweiler, and Saint Bernard.

¶ Airedale Terrier, Cocker Spaniel, Dachshund, Doberman Pinscher, Golden Retriever, Irish Setter, Miniature Schnauzer, Pomeranian, and Shetland Sheepdog.

[‡] Boxer, English Setter, German Shepherd Dog, Golden Retriever, Great Dane, Labrador Retriever, Pointer, Poodle, and Siberian Husky.

[§] Akita, American Staffordshire Terrier, Chesapeake Bay Retriever, German Shepherd Dog, Golden Retriever, Labrador Retriever, Mastiff, Neapolitan Mastiff, Newfoundland, Poodle, and Saint Bernard.

II Beagle, Cairn Terrier, Cavalier King Charles Spaniel, Cocker Spaniel, Dachshund, Labrador Retriever.

Does Zeuterin[™] Change Behavior?

Behavior changes may be observed after zinc neutering:

"My observation as an Animal Caretaker for the dogs is that the male dogs treated with Zinc Gluconate are much calmer than as compared to the intact male dogs." – FDA Clinical Trial, Progeny Testing and Sexual Behavior Study

"Max stopped urine marking and now sits on my lap" – Dee Ann, Dog Owner

"Tank stopped digging the garden and became cuddly" – Kari, Dog Owner

What We Think We Know

(Quotes from websites of veterinary clinics, humane societies, trainers & animal shelters

"Spaying and neutering makes pets better, more affectionate companions." "Unsterilized animals often exhibit more behavior and temperament problems than do those who have been spayed or neutered."

"Female dogs, like males, have an increased risk of aggression if left intact."

"Unneutered dogs are often more aggressive and territorial (urine marking, fighting), but these traits should not be confused with loyalty and protection of their home and family."

"Altered pets are less aggressive toward other dogs and cats, are less likely to urine mark and wander, and generally have better personalities."

Behavior Conclusions

- Surgical neutering may reduce specific male hormonedependent behaviors (e.g. sexual mounting, roaming, urine-marking, and aggression directed toward other (intact) males) in dogs that have already learned these behaviors.
- Surgical spay/neuter may increase other undesirable behaviors (e.g. owner-directed aggression, touch sensitivity, fearfulness, etc.) in otherwise behaviorally normal dogs.
- Definitive conclusions will require prospective, controlled studies.

Does Zeuterin[™] Change Behavior?

The FDA behavior position statement:

"As with surgical castration, secondary male characteristics (roaming, marking, aggression, or mounting) may be displayed."



Zeuterin Administration

Measure the testicular width

Gently cleanse and disinfect the scrotum



Courtesy Dr. Brenda Griffin

Zeuterin Administration

- Pass the needle from the cranial aspect of the testicle, just ventral to the head of the epididymis
- Position the needle along the long axis of the testicle in the center of the testicular parenchyma



Zeuterin vs. Neutersol What has changed?

Injection technique modified – strict adherence to protocol is important to minimize potential adverse reactions

 \checkmark Three 28 gauge needles, ½ to 1 inch length

- \checkmark 1 needle to draw solution 2 needles to inject
- \checkmark Timed injection (slow)

Zeuterin vs. Neutersol What has changed?

Injection technique modified – strict adherence to protocol is important to minimize potential adverse reactions

- \checkmark Do not aspirate prior to injecting
- \checkmark Wait when done injecting prior to withdrawal
- \checkmark Do not massage the testicles after injecting

How Do We Identify a Zinc Neutered Dog?

Microchipping

 Microchip tracking companies will record the Zinc Neutered status of dog

Tattooing

- A "Z" tattoo between the genitals and inner thigh is the standard sign for zinc neutering Unique Collar Tag
 - Every dog is given a unique identification number collar tag with the website address to verify neutering status

Changing Perception

Evidence-based decision making



- Conscientious, explicit, and judicious use of current best evidence in making decisions about the care of individual patients.
- De-emphasizes intuition, unsystematic clinical experience, and pathophysiologic rationale as sufficient grounds for clinical decision making and stresses the examination of evidence from clinical research.

Changing Perception



When I was in vet school, I was told, "Half of what you learn here will turn out to be wrong. But no one knows which half."

So true! One of the things we may have gotten wrong was in looking at the health impacts of neutering.

Dr Marty Becker "America's Veterinarian"

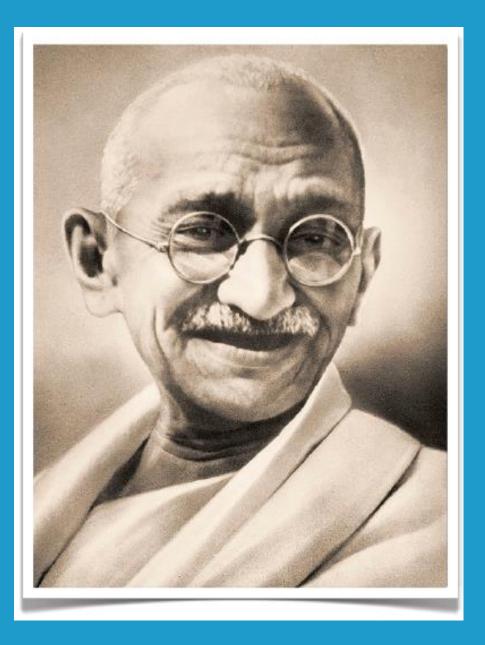
The Evolution of Neutering



Zinc Gluconate Neutralized by L-Arginine is the only FDA Approved Non-surgical Sterilant



A single, virtually painless injection into each testicle will sterilize a male dog, permanently.



"The greatness of a nation and its moral progress can be judged by the way its animals are treated."

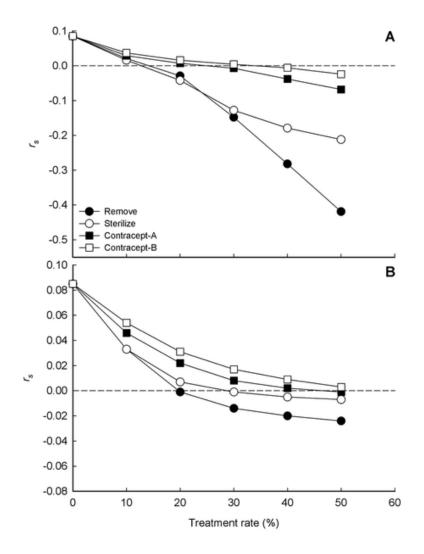
Mahatma Gandhi

Projects to pave the way...



Population Models for Free-Roaming Cats

Miller PS, Boone JD, Briggs JR, Lawler DF, Levy JK, et al. (2014) Simulating Free-Roaming Cat Population Management Options in Open Demographic Environments. PLoS ONE 9(11): e113553. doi:10.1371/journal.pone.0113553 Figure 6. Stochastic population growth rate under different FRC management strategies.



Miller PS, Boone JD, Briggs JR, Lawler DF, et al. (2014) Simulating Free-Roaming Cat Population Management Options in Open Demographic Environments. PLoS ONE 9(11): e113553. doi:10.1371/journal.pone.0113553 http://www.plosone.org/article/info:doi/10.1371/journal.pone.0113553



Marking and identification of freeroaming populations of dogs and cats





Cornell University David R. Atkinson Center for a Sustainable Future

International Society of Feline Medicine 2015 Feline Veterinary Congress

Pre-Congress Day: "Feline Fertility and Population Control"
Special issue of Journal of Feline Medicine and Surgery





acc-d.org

- ✓ E-book
- Product profile & position papers

Contraception and

Fertility Control in

Dogs and Cats

A Report of the Alliance for Contraception in Cats & Dogs (ACC&D)

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- Legislative information
- FAQs
- ✓ Much more

Thank you for exploring the options with us.



acc-d.org







imagine... preventing unwanted litters without surgery



Surgery Not Required: Current and Future Options in Fertility Control of Dogs and Cats

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