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## Proceedings of the Vertebrate Pest Conference

### Title

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### Permalink

<https://escholarship.org/uc/item/2xr915qh>

### Journal

Proceedings of the Vertebrate Pest Conference, 31(31)

### ISSN

0507-6773

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### Publication Date

2024

# Efficacy of GonaCon Fertility Control against Prairie Dogs and Potential for Uses on Other Rodent Species

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**ABSTRACT:** Use of a contraceptive (i.e., fertility control) is attractive for rodent management where lethal control is unwanted. Although population reduction is generally unachievable with small-scale, short-term contraceptive use, reduced juvenile recruitment is achievable. The injectable immunocontraceptive vaccine GonaCon (active ingredient: gonadotropin releasing hormone [GnRH]) was registered in 2022 by the U.S. EPA for controlling fertility of female prairie dogs (*Cynomys*, a type of ground squirrel) in urban/suburban settings. Here we: 1) describe past research, including a replicated field study in Colorado (GonaCon treatment vs. control sites) testing efficacy of GonaCon in prairie dogs, which gave rise to the EPA registration of this product, 2) outline future research needs for prairie dog population management with GonaCon–Prairie Dogs, 3) describe the steps required to possibly register GonaCon for additional rodent species, and 4) describe the other fertility control pesticide products that are currently registered for use against rodents in the U.S. During the replicated field study in Colorado, prairie dogs were live-trapped in a portion (avg: 18.5%, range: 7-37%) of each colony's total area. In treatment plots, every female captured  $\geq 660$  g was injected with 0.4 ml of GonaCon. GonaCon was highly effective in controlling female fertility during the first year (2019), as juvenile density was reduced 3 $\times$ , but not in the second year (2020) following treatment. Treating whole colonies of prairie dogs is favored, yet if small or partial colony treatment is desired then annual GonaCon treatment may be needed. An amendment to the EPA label is in progress to allow treatment of both male and female prairie dogs. Due to recent interest from land and pest managers, other fertility control products and the steps required to possibly get GonaCon registered for additional rodent species are also outlined.

**KEY WORDS:** animal population management, contraceptive injection, *Cynomys* spp., ground squirrel, non-lethal fertility control, prairie dog, vaccine

Proceedings, 31<sup>st</sup> Vertebrate Pest Conference (R. M. Timm and D. M. Woods, Eds.)

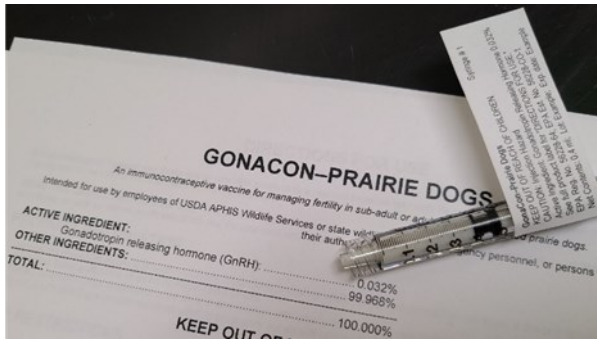
Paper No. 6. Published August 30, 2024. 5 pp.

## INTRODUCTION

One category of non-lethal control for prairie dog management is the use of contraceptive pesticide products (i.e., substances that control fertility of a pest species). Although population reduction is generally unachievable with small-scale, short-term contraceptive use in prairie dog colonies where immigration transpires, reduced production of juveniles is achievable. The injectable immunocontraceptive vaccine GonaCon (active ingredient is Gonadotropin releasing hormone [GnRH] at 0.032% w/w) is an effective contraceptive for many mammal species (Miller et al. 2013). GonaCon works as follows. GnRH is a peptide hormone produced by the hypothalamus, and it is a key hormone in the control of reproduction in mammals. When an animal is injected with GonaCon, it stimulates an immune response where antibodies against GnRH are produced and thus bioactivity of GnRH is suppressed. This in turn suppresses the production of other reproductive hormones, including gonadal steroids, resulting in decreased fertility. Identical formulations of GonaCon are currently federally registered with the U.S. Environmental Protection Agency (EPA) under Section 3 (hereafter, referred to as a Section 3 registration) of the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) for use on female feral or wild equine (feral or wild horses [*Equus ferus caballus*] and burros [*E. asinus*], product names GonaCon–Equine, EPA Reg. No. 56228-41, and GonaCon EQ, EPA Reg. No. 92105-1) and female white-tailed deer (*Odocoileus virginianus*; product name GonaCon–Deer; EPA Reg. No. 56228-40). In 2022, the U.S. Department of Agriculture's (USDA) Animal and

Plant Health Inspection Service (APHIS) registered the same formulation as GonaCon–Prairie Dogs with EPA for use on three species of prairie dogs, which are a type of ground squirrel (black-tailed prairie dog, *Cynomys ludovicianus*; white-tailed prairie dog, *C. leucurus*; and Gunnison prairie dog, *C. gunnisoni*; EPA Reg. No. 56228-64) in urban/suburban settings, open spaces and natural areas, parks, campgrounds, airports, roadway medians, and other non-crop use sites. GonaCon–Prairie Dogs is not labeled for use on prairie dog colonies solely located within cropland, pastures, or rangeland used for grazing or animal feed production. This avoidance of use within agricultural settings was likely a result of the agricultural industry needing rapid population reduction or eradication of prairie dogs, as prairie dogs are well-known for their crop damage and competition with grazing livestock (Collins et al. 1984, Hygnstrom and Virchow 1994), as well as carriers of plague (Witmer et al. 2023). GonaCon–Prairie Dogs can only be applied by APHIS-Wildlife Services (WS) or state wildlife agency personnel or persons under their authority. GonaCon–Prairie Dogs is currently registered in two states, Colorado and New Mexico.

The following characteristics make GonaCon–Prairie Dogs an attractive non-lethal tool for prairie dog management: 1) it must be injected (Figure 1) and therefore, has zero primary risks to non-target species, 2) GnRH is rapidly metabolized by mammals and quickly denatures in both the natural environment and in the digestive tract of



**Figure 1. GonaCon–Prairie Dogs is a 0.4 ml dose that arrives in a 1 ml syringe. It does not require mixing, but arrives cold and should be maintained at 5°C (i.e., refrigerator temperature) until it is injected into the prairie dog. It should not be frozen. It is ordered from USDA/APHIS/Wildlife Services.**

birds and mammals and therefore it has very low to zero secondary risks to non-target species (EPA 2020), 3) it is estimated to be a lifetime vaccine for most wild prairie dogs due to their short lifespans (i.e., assumed effectiveness for 3 years; Yoder et al. 2008), and 4) it is relatively affordable (currently ~\$18/individual dose) (Shiels et al. 2024). Land managers in California and other states have expressed interest in non-lethal contraceptive products such as GonaCon for use in ground squirrels (e.g., California ground squirrels, *Otospermophilus beecheyi*), tree squirrels (e.g., *Sciurus niger*, *S. carolinensis*), and possibly other rodents (e.g., *Rattus*, *Mus*, *Microtis*, *Peromyscus*, Geomyoidea).

In this paper, we 1) review the product performance studies that supported the recent Section 3 registration of GonaCon–Prairie Dogs, 2) outline future research needs for prairie dog population management with GonaCon–Prairie Dogs, 3) describe the steps required to possibly register GonaCon for additional rodent species, and 4) provide a listing of the other fertility control pesticide products that are currently registered for use against rodents in the U.S..

### **PAST RESEARCH SUPPORTING GONACON– PRAIRIE DOGS EPA REGISTRATION**

USDA’s National Wildlife Research Center (NWRC) began immunocontraceptive vaccine research for wildlife damage management in 1992, beginning with testing porcine zona pellucida (PZP) in white-tailed deer (Miller et al. 2013). Suppression of GnRH was quickly realized as an efficient pathway to reduce fertility in both sexes of deer, but with greatest effectiveness in females and suppression of GnRH in males having the potential to disrupt antler formation. An appropriate and economically viable protein carrier for the GnRH peptide was identified as blue protein, derived from a mollusk (*Concholepas concholepas*), and this carrier protein is now used in GonaCon products (Miller et al. 2013). GonaCon–Prairie Dogs (0.032% w/w GnRH) is administered as an intramuscular injection that can achieve contraception in individual prairie dogs for multiple years even after a single injection (Yoder et al. 2008).

Past product performance research with GonaCon and prairie dogs included both a laboratory trial (Yoder and Miller 2011) and a small-scale field efficacy trial without plot replication (Yoder 2011). These trials showed GonaCon had high efficacy in limiting female prairie dog reproduction as well as some male reproduction inhibition. Specifically, Yoder and Miller (2011) conducted a laboratory trial with prairie dogs that demonstrated that 0.2 ml or 0.4 ml injections of GonaCon produced ample titers that would likely contracept prairie dogs for at least 15 months. In a field study using live-trapping with one GonaCon treatment colony and one control colony, Yoder (2011) reported that none (0%) of the animals treated with GonaCon and recaptured a year later were breeding compared to 94.4% of animals administered a sham vaccine (absent of GnRH) and recaptured a year later in the control site. In addition, in the two years following treatment, the ratio of pups to adults from visual counts averaged 79% lower in the GonaCon-treated site compared to the control site.

In attempt to build on the Yoder and Miller studies and to support a Section 3 application for EPA registration of GonaCon for use with prairie dogs, a replicated field product performance study was conducted in prairie dog colonies in Fort Collins and Denver, Colorado, under an Experimental Use Permit (EUP) from EPA (EUP No. 56228-EUP-44) and the Colorado Department of Agriculture. The details of this study can be found in Shiels et al. (2024). Briefly, prairie dogs were live-trapped in October–December 2018 in three treatment and three control sites. Only a portion (avg: 18.5%, range: 7-37%) of each colony’s total area was trapped. Every female captured with  $\geq 660$  g bodyweight was injected with 0.4 ml of either GonaCon at treatment sites or a sham vaccine at control sites. 23-gauge stainless steel hypodermic needles were used for each injection. Juveniles and adults were counted at all six sites to establish juvenile: adult (ratio) and juvenile density in May–June 2019 (determining initial GonaCon effectiveness) and 2020 (determining persistence of contraception). In 2019, the juvenile to adult ratio was significantly reduced ( $P = 0.0022$ ) in GonaCon (mean  $\pm$  SE:  $0.23 \pm 0.01$ ) relative to the sham ( $1.10 \pm 0.58$ ) sites. Juvenile density was three times greater in sham than GonaCon sites. In 2020, GonaCon sites had similar juvenile densities as nearby sham sites. The main conclusions from the study were that: 1) GonaCon is highly effective at reducing juvenile recruitment in a colony during the first year (2019) but not in the second year (2020) if only a small area (0.6–2.8 ha) or small proportion of females in a colony is treated like in this study, and 2) treating a whole colony of prairie dogs at once is recommended for greater population control, but if it is possible to only treat part of a colony, then annual (repeated) GonaCon treatments of that colony may be needed to maintain lower juvenile recruitment over time (Shiels et al. 2024).

In early 2024, APHIS submitted a label amendment application to EPA to add the option to treat males based on results from the Yoder and Miller studies, as currently only females can be treated. EPA’s decision on the label amendment is expected in mid-2024.

## FUTURE RESEARCH NEEDS FOR GONACON–PRAIRIE DOGS

In 2024, Drs. Shiels and Bruemmer proposed a multi-agency prairie dog study in Colorado’s Front Range that will 1) treat all males and females, if allowed under an amended label, 2) treat whole colonies, and 3) include the following three treatments to better guide land managers with prairie dog colonies within allowed use sites on the label: administer GonaCon–Prairie Dogs only, cull 75% of the colony+administer GonaCon–Prairie Dogs to the survivors, and cull 75% of the colony only. Reference sites (i.e., “controls”) where no culling, trapping, or GonaCon–Prairie Dogs use will occur will be established at each field site. Once treatments occur (October 2024), offspring production and population size changes will be measured for three seasons (i.e., June 2025, 2026, 2027). This study will advance data sharing and identify optimal treatment effectiveness that each agency may consider for future prairie dog colony management.

Additional future research involving prairie dogs and GonaCon–Prairie Dogs may include additional NWRC laboratory and pen studies to identify the ideal dosage, efficacy in males vs. females, and the duration of reproductive inhibition. Other oral contraceptive baits that are not currently registered by the EPA also may be tested using the experimental prairie dog colony planned for establishment at NWRC.

## REQUIREMENTS FOR GONACON USE WITH OTHER RODENT SPECIES

Land managers in California and other states have expressed interest in using GonaCon for ground squirrels (e.g., California ground squirrels, *Otospermophilus beecheyi*), tree squirrels (e.g., *Sciurus niger*, *S. carolinensis*), and possibly other pest rodent species (e.g., *Rattus*, *Mus*, *Microtis*, *Peromyscus*, Geomyoidea). Here we provide a brief overview of the regulatory data submissions (registration studies) likely to be required by EPA and some state regulatory agencies (e.g., California Department of Pesticide Regulation (CDPR); [https://www.cdpr.ca.gov/docs/registration/data\\_requirements.pdf](https://www.cdpr.ca.gov/docs/registration/data_requirements.pdf)) in future applications for EUPs and to register the current GonaCon formulation for additional target rodent species and use patterns.

EPA’s data requirements for EUP and Section 3 registration applications, including label amendment applications, for different categories of pesticides and use patterns are listed in 40 CFR 158. The corresponding test guidelines that these registration studies must follow are available at <https://www.epa.gov/test-guidelines-pesticides-and-toxic-substances/final-test-guidelines-pesticides-and-toxic>.

Registration studies that are submitted to EPA to support an EUP approval or registration action must be conducted in accordance with EPA’s FIFRA Good Laboratory Practice (GLP) regulations outlined in 40 CFR 160.

For a vertebrate contraceptive product with a terrestrial outdoor, non-food crop use pattern, these data requirements fall under the following EPA data categories in 40 CFR 158: product chemistry (product properties), toxicology (human health effects), ecological effects, environmental fate, and product performance. The majority of these product chemistry, toxicology, ecological effects, and

environmental fate registration studies must be conducted on the technical grade of the active ingredient (TGAI; e.g., GnRH without any inert ingredients added other than those used for purification of the active ingredient). The “Group A” product chemistry studies (40 CFR 158.310–158.355; <https://www.epa.gov/test-guidelines-pesticides-and-toxic-substances/series-830-product-properties-test-guidelines>) are required for each proposed manufacturing source of the active ingredient and end-use product (e.g., GonaCon–Prairie Dogs). A subset of the “Group B” product chemistry (physical and chemical properties; 40 CFR 158.310) and toxicology studies (40 CFR 158.500) must also be submitted for the end-use product. EPA then uses these data to evaluate a product’s consistency and quality, and to assess risks to humans, domestic animals, wildlife, and the environment when making registration decisions for pesticides (40 CFR 158.130; <https://www.epa.gov/pesticide-registration/data-requirements-pesticide-registration>).

EPA and some state regulatory agencies like CDPR also require that field product performance studies are submitted in a Section 3 registration application for pesticides targeting vertebrates designated as public health pests and on a case-by-case basis for any pesticide product registered or proposed for registration (40 CFR 158.400(e)). EPA’s list of public health pests is available at <https://www.epa.gov/insect-repellents/list-pests-significant-public-health-importance>. EUPs are issued by EPA and some state pesticide regulatory agencies to allow these field studies to be conducted in locations and under conditions representative of the proposed future operational use patterns (40 CFR 172). The potential exception is when additional target rodent species and use patterns are similar enough to those already registered for use that EPA does not require additional product performance studies. This can be determined through consultation with EPA. However, even if EPA does not require new product performance studies, some states like California may still require them for state registration of the product or use for that target species.

Within an EUP application to EPA, the registrant submits a draft EUP field study protocol that proposes how the performance of the end-use product will be tested. The EUP application must also include submission (or citation) of many of the data requirements for the TGAI and end-use product described above. EPA uses these data to evaluate the product’s composition and any potential risks of the proposed EUP study. For public health pests, the registrant also submits or cites a laboratory efficacy (product performance) study conducted with the end-use product and the proposed target species (or appropriate proxy species) and dosage. EPA uses the results of the laboratory efficacy study to evaluate whether the product’s efficacy warrants further testing in the field in the proposed EUP field study. To help ensure that EPA will accept the laboratory efficacy study results to support the EUP, the proposed laboratory efficacy study protocol can be submitted to EPA before conducting the study for EPA’s review and any requests for changes.

When approving an EUP, EPA will specify the conditions the EUP field study must follow and any additional recommended changes to the EUP field study protocol. Once the EUP is approved by EPA, the EUP and proposed

study details are submitted to the state pesticide regulatory agency in the state(s) in which the field study will be conducted for their authorization or registration, if required by state law. Once the EUP field study is authorized by the state(s), the field product performance study must then be conducted in accordance with the EPA-approved protocol and FIFRA GLPs. If the product performs well in the EUP study, the registrant submits the study to EPA in a new Section 3 registration or label amendment application allowing the new use along with the other required data.

Under the potential scenario where APHIS is the future GonaCon product registrant and manufacturer, adding a new target rodent species could be accomplished with a new Section 3 product registration application or by potentially amending the current GonaCon–Prairie Dogs registration with EPA to allow additional rodent target species and use instructions. In most cases, these registration applications would require submittal of new product performance data, including a completed EUP field study as described above. Given the set of registration studies for GnRH and the GonaCon end-use product formulation that APHIS has already submitted to and accepted by EPA to date, the only additional registration study that EPA would likely require to support an EUP application is a new laboratory efficacy (product performance) study. The completed EUP study along with a data matrix citing the existing registration data for GnRH and GonaCon could then be submitted to EPA in a new Section 3 registration or label amendment application.

Under a scenario where another company was the registrant and manufacturer of a new GonaCon product for additional target rodent species, then additional registration studies must be completed and submitted in an application for an EUP (and a future Section 3 registration). First, the new manufacturer of the GonaCon product would have to complete the Group A product chemistry studies for their end-use product. Ideally, the laboratory efficacy study would also test their GonaCon product made at the new manufacturing site. If a new manufacturing source of GnRH was used in their GonaCon product, then new Group A product chemistry data would also be required on this new manufacturing source of GnRH. The data requirements for applying for the new Section 3 registration after the EUP was completed would then be the same as described above for the scenario where APHIS is the registrant.

#### **ADDITIONAL FERTILITY CONTROL PRODUCTS DEVELOPED**

In addition to GonaCon–Prairie Dogs, there are two other fertility control products currently available for use against rodents in the U.S., and both are from SenesTech<sup>®</sup>, Inc. (Phoenix, Arizona): 1) ContraPest<sup>®</sup> is a federally registered (FIFRA, Section 3 registration; EPA Reg. No. 91601-1) contraceptive product for control of non-native *Rattus* spp. in or immediately surrounding man-made structures and must be applied in tamper-resistant bait stations. ContraPest is an oral liquid bait that contains the active ingredients 4-vinylcyclohexane diepoxide (VCD; 0.09604 % w/w) and triptolide (0.00118 % w/w). ContraPest was also tested by the NWRC in published laboratory efficacy trials for both Norway rats (*R. norvegicus*; Witmer

et al. 2017) and black rats (*R. rattus*; Siers et al. 2020). 2) Evolve<sup>™</sup> is a FIFRA Section 25(b) minimum risk pesticide (MRP) contraceptive product, therefore exempt from further federal registration (and not reviewed by EPA in any capacity), and it is currently available for control of non-native *Rattus* spp. (although additional rodents may be added in the future) in most states and in a variety of use sites, including natural areas. Evolve is a soft bait, and the minimum risk pesticide active ingredient is cottonseed oil (0.1% w/w), which may contain a compound called gossypol. Both ContraPest and Evolve are recommended to be continuously delivered in bait stations for control, because when rats stop eating these products (i.e., triptolide for ContraPest) they regain fertility (e.g., in approximately 6 weeks for Evolve, <https://go.senestech.com/evolve-rats-faq#page=1&navpanes=0&view=fitH,100>). The second active ingredient in ContraPest, VCD, causes irreversible sterility in females due to total follicle loss. As described in Witmer et al. (2017), VCD-induced follicle loss occurred within 15 days of daily dosing, and an average of 46 days after cessation of the dosing to reach complete ovarian failure. Consumption of ContraPest and Evolve by nontarget species (e.g., birds and mammals other than target rats) may cause infertility, which is again why bait stations are required or recommended for the delivery of these baits. Neither product has been tested against prairie dogs or other ground squirrels to our knowledge, so such efficacy testing in the laboratory would be a first step if these products were to be considered for prairie dogs or other species.

Although not registered with EPA, DiazaCon (synonyms: Azacosterol hydrochloride; desmosterol; 20,25-Diazacholesterol dihydrochloride), a cholesterol inhibitor, is a promising contraceptive active ingredient for rodents and birds, and that has been previously tested in oral baits with prairie dogs (Yoder et al. 2016) and parrots (Lambert et al. 2010). Oral bait contraceptive products greatly reduce the time and labor requirements for application for target rodent species compared to products that require capture and intramuscular injection of the target animals. However, unlike injectable products, oral baits have potential non-target species primary and secondary exposure risks and there is no certainty that each target animal has been exposed with an effective dosage. DiazaCon administered in oral baits has been shown to be highly effective at reducing fertility in black-tailed prairie dogs in Colorado. Specifically, when a single treatment plot and a single control plot were compared, DiazaCon baits reduced the number of pups per adult by 95% (Yoder et al. 2016). Another potential drawback of using DiazaCon is that it must be fed to prairie dogs over multiple days prior to each breeding season to reduce annual fecundity (Yoder et al. 2016).

As far as we know, DiazaCon is not currently being pursued for EPA registration for prairie dogs or other species, and this is largely due to the scarcity and formidable expense of the active ingredient and registration data costs. If it were to be pursued further in the future, DiazaCon baits would have to be used in bait stations because prairie dog colonies and other target ground and tree squirrel populations have resident and visitor nontarget wildlife species, such as native birds (e.g., corvids,

passerines, Canada geese, raptors) and mammals (e.g., mice, skunks, rabbits, foxes, coyotes, deer), which may be attracted to the baits themselves or secondarily exposed.

It is more difficult for us to thoroughly review the other potential fertility control products that are registered for use against rodents outside the U.S. However, there is one registered in Tanzania against multimammate mice (*Mastomys natalensis*), and it is referred to as EP-1. Two hormones (levonorgestrel and quinestrol) comprise the active ingredients of EP-1 (Shi et al. 2020, Massei et al. 2024).

## ACKNOWLEDGEMENTS

Mention of a company or commercial product does not mean endorsement by the U.S. government.

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