Date of Approval: February 20, 2026

FREEDOM OF INFORMATION (FOI) SUMMARY

APPLICATION FOR CONDITIONAL APPROVAL

Application number 141-617

Exzolt™ Cattle-CA1

(fluralaner topical solution)

Beef cattle 2 months of age and older and replacement dairy heifers less than 20 months of age

Exzolt™ Cattle-CA1 is indicated for the prevention and treatment of infestations caused by New World screwworm (*Cochliomyia hominivorax*) larvae (myiasis) and treatment and control of cattle fever tick (*Rhipicephalus microplus*) in beef cattle 2 months of age and older and replacement dairy heifers less than 20 months of age.

Sponsored by:

Intervet, Inc.

Executive Summary

Exzolt™ Cattle-CA1 (fluralaner topical solution) is conditionally approved for the prevention and treatment of infestations caused by New World screwworm (*Cochliomyia hominivorax*) larvae (myiasis) and treatment and control of cattle fever tick (*Rhipicephalus microplus*) in beef cattle 2 months of age and older and replacement dairy heifers less than 20 months of age.

The Food and Drug Administration (FDA) determined that Exzolt™ Cattle-CA1 is eligible for conditional approval for the labeled use (prevention and treatment of infestations caused by New World screwworm (*Cochliomyia hominivorax*) larvae (myiasis)) under section 571(a)(1)(A)(ii) of the Federal Food, Drug, and Cosmetic Act (FD&C Act) because the drug prevents and/or treats a serious or life-threatening disease in cattle and demonstrating effectiveness would require complex or particularly difficult studies.

The FDA determined that Exzolt™ Cattle-CA1 is eligible for conditional approval for the labeled use (treatment and control of cattle fever tick (*Rhipicephalus microplus*)) under section 571(a)(1)(A)(ii) of the FD&C Act because the drug fulfills an unmet animal need as there are currently no FDA-approved drugs with an indication for cattle fever tick. Additionally, for this indication, demonstrating effectiveness would require complex or particularly difficult studies.

A conditionally approved animal drug has been shown to be safe and has a reasonable expectation of effectiveness. During the conditional approval period, the sponsor can legally market the drug for the labeled use while collecting the remaining effectiveness data. The conditional approval is valid for one year. The sponsor can ask FDA to renew the conditional approval annually for up to four more years, for a total of five years of conditional approval. To receive a renewal from FDA, the sponsor must show active progress toward proving substantial evidence of effectiveness for full approval.

For the conditional approval of the New World screwworm (NWS) indication, three study reports utilizing natural infestations of *C. hominivorax* were used to support reasonable expectation of effectiveness. These studies were conducted in Brazil in 2018.

For the conditional approval of the cattle fever tick indication, a total of 17 study reports were used to support reasonable expectation of effectiveness: 13 were field effectiveness studies utilizing natural infestations of *R. microplus*, 3 were dose confirmation studies utilizing induced infestations, and 1 was a rain exposure study utilizing induced infestation. For more information on these studies, see the Effectiveness section below.

FDA concluded these studies were acceptable to support reasonable expectation of effectiveness for conditional approval for both NWS and cattle fever tick indications.

The target animal safety of Exzolt™ Cattle-CA1 was evaluated in two studies: one margin of safety study and one female cattle reproductive safety study. In the margin of safety study, Exzolt™ Cattle-CA1 was administered to healthy beef cattle at up to five times the labeled dose and three times the treatment duration. No clinically significant adverse effects were observed. The female reproductive safety study in beef cattle

administered three times the labeled dose and showed no adverse effects on reproductive performance or offspring viability.

A human food safety evaluation of Exzolt™ Cattle-CA1 was conducted. Based on toxicology studies, the acceptable daily intake for total residue of fluralaner was established as 10 µg/kg body weight/day. The calculated safe concentrations for total residues of fluralaner were 440 parts per billion (ppb) in muscle, 1,320 ppb in liver, 2,640 ppb in kidney, and 2,640 ppb in fat. Residue depletion studies determined that cattle liver is the target tissue and that parent fluralaner is the marker residue for monitoring residues in cattle liver and muscle. Tolerances of 500 ppb in cattle liver and 350 ppb in cattle muscle were established. Environmental temperature was determined to affect the withdrawal period. Cattle must not be slaughtered for human consumption within **98 days** of treatment. If cattle are continuously exposed to temperatures at or above 60 °F after product administration, then cattle may be slaughtered for human consumption 44 days after treatment. Violative residues may result if cattle are exposed to temperatures below 60 °F after administration and are slaughtered at 44 days. As this drug is not an antimicrobial, FDA determined that a microbial food safety assessment was not required.

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I. GENERAL INFORMATION

A. File Number

Application number 141-617

B. Sponsor

Intervet, Inc.
126 E Lincoln Ave.
Rahway, NJ 07065

Drug Labeler Code: 000061

C. Proprietary Name

Exzolt™ Cattle-CA1

D. Drug Product Established Name

fluralaner topical solution

E. Pharmacological Category

Antiparasitic

F. Dosage Form

Solution

G. Amount of Active Ingredient

50 mg fluralaner/mL

H. How Supplied

250 mL, 1L, and 5L bottles

I. Dispensing Status

Prescription (Rx)

J. Dosage Regimen

Exzolt™ Cattle-CA1 is a ready-to-use topical formulation intended for direct application to the hair and skin in a narrow strip extending along the dorsal midline from the withers to the base of the tail. The recommended rate of administration is 1 mL/44.1 lbs. (1 mL/20 kg) body weight, which is equivalent to 1.13 mg of fluralaner for each pound (2.5 mg/kg) body weight.

K. Route of Administration

Topical

L. Species/Classes

Beef cattle 2 months of age and older and replacement dairy heifers less than 20 months of age

M. Indication

Exzolt™ Cattle-CA1 is indicated for the prevention and treatment of infestations caused by New World screwworm (*Cochliomyia hominivorax*) larvae (myiasis) and treatment and control of cattle fever tick (*Rhipicephalus microplus*) in beef cattle 2 months of age and older and replacement dairy heifers less than 20 months of age.

II. EFFECTIVENESS

Conditional Dose: The conditional dose for the indication "for the prevention and treatment of infestations caused by New World screwworm (*Cochliomyia hominivorax*) larvae (myiasis) and treatment and control of cattle fever tick (*Rhipicephalus microplus*) in beef cattle 2 months of age and older and replacement dairy heifers less than 20 months of age" is 2.5 mg/kg body weight (1 mL/20 kg) once topically along the dorsal midline from withers to the base of the tail. The safety data and the data to demonstrate reasonable expectation of effectiveness provide support for this conditional dose.

A. Dosage Characterization

Based on effectiveness data generated in dose determination studies with Exzolt™ Cattle-CA1 (fluralaner topical solution) against the single-host tick *Rhipicephalus microplus*, an appropriate effective dose was determined to be 2.5 mg fluralaner/kg body weight in beef cattle.

B. Reasonable Expectation of Effectiveness

1. Reasonable expectation of effectiveness for Exzolt™ Cattle-CA1 (fluralaner topical solution) for the prevention and treatment of infestations caused by New World screwworm (*Cochliomyia hominivorax*) larvae (myiasis) was based on three effectiveness studies utilizing natural NWS infestations conducted in Brazil in 2018:

    a. Support for a prevention indication: This study evaluated prevention of New World screwworm (NWS) myiasis in a surgical wound created seven days after treatment administration. Animals received either a placebo (n=6) or Exzolt™ Cattle-CA1 (n=6) on Day -7. Seven days later, two surgical incisions were made on each side of the body at the shoulder. Animals were housed outside to facilitate natural infestation of the wounds with NWS. Cattle were monitored twice daily for 10 days post-incision to assess the presence of eggs and larvae. A single topical application of Exzolt™ Cattle-CA1 at the dose of 2.5 mg/kg provided 100% prevention against myiasis for the length of the study.

    b. Support for a prevention indication: This study evaluated prevention of NWS myiasis in a castration wound created on the day of treatment with either a placebo (n=15) or Exzolt™ Cattle-CA1 (n=15). Animals were housed outside

to facilitate natural infestation of the wounds with NWS. Cattle were monitored daily for 14 days post-surgery to assess the presence of eggs, larvae, and the progress of wound healing. A single topical administration of Exzolt™ Cattle-CA1 at the dose of 2.5 mg/kg provided 100% prevention against myiasis for up to 14 days following castration.

c. Support for a therapeutic indication: This study evaluated the effectiveness of the product to treat a wound already infested with NWS. A surgical wound was created and left exposed to facilitate natural infestation with NWS. Three days later, after confirming the presence of live larvae, animals were treated topically once with either a placebo (n=12) or Exzolt™ Cattle-CA1 (n=12). A single topical administration of Exzolt™ Cattle-CA1 at the dose of 2.5 mg/kg achieved 90.9% effectiveness by the second day post-treatment and reached 100% effectiveness by the third day. No myiasis in treated animals was observed up to day 5.

2. Reasonable expectation of effectiveness for Exzolt™ Cattle-CA1 (fluralaner topical solution) for the treatment and control of cattle fever tick (*Rhipicephalus microplus*) was based on a total of 17 studies as summarized below. Length of ≥ 99% efficacy is reported here because *R. microplus* is a reportable tick in the United States and drugs used for its treatment and control must have a very high efficacy.

Table II.1. Studies Supporting Reasonable Expectation of Effectiveness

Study Number	Study/Infestation Type	Study Location	Animals/Group	Percent Efficacy
S20218-00	Field study/natural infestation	Australia	22	100% Days 3 - 70
S20218-01	Field study/natural infestation	Australia	22	100% Days 5 - 58
S20218-02	Field study/natural infestation	Australia	22	100% Days 3 - 49
S20218-03	Field study/natural infestation	Australia	22	100% Days 3 - 50
S20218-04	Field study/natural infestation	Australia	22	100% Days 3 - 49
S20218-05	Field study/natural infestation	Australia	22	100% Days 7 - 49
S17258	Field study/natural infestation	Brazil	10	100% Days 7 - 42
S17259	Field study/natural infestation	Brazil	10	100% Days 3 - 56

Study Number	Study/Infestation Type	Study Location	Animals/Group	Percent Efficacy
S17260	Field study/natural infestation	Brazil	10	100% Days 21 - 35
S18280	Field study/natural infestation	Brazil	10	100% Days 7 – 35
S18281	Field study/natural infestation	Brazil	10	100% Days 11 - 28
S17175-01	Field study/natural infestation	Australia	20	100% Days 4 – 50
S17175-03	Field study/natural infestation	Australia	20	100% Days 7 – 58
S19152	Rain exposure study/induced infestation	Brazil	6	100% Days 7 - 77
S17261	Dose confirmation study/induced infestation	Brazil	8	100% Days 8 -110
S17262	Dose confirmation study/induced infestation	Brazil	8	100% Days 6 – 37

Study Number	Study/Infestation Type	Study Location	Animals/Group	Percent Efficacy
S19173	Dose confirmation study/induced infestation	South Africa	4 (4 treated animals were not allowed to lick for 16 days; 4 treated animals were allowed to lick)	Animals not allowed to lick for 16 days did not reach >95% effectiveness during that time. Once allowed to lick, efficacy rose to 100% by Day 18 and was maintained until Day 39. Animals allowed to lick for 10 days post-treatment achieved 96.5% efficacy on Day 39, which was when the corresponding control group reached adequate infestation levels. Efficacy fell below 80% on Days 48 and 49.

Out of the 17 studies listed above, 3 dose confirmation studies conducted in Brazil and South Africa utilizing induced infestations of *R. microplus* were evaluated. These studies were conducted between 2018 and 2021. In each study, animals were individually housed and randomly assigned to control and Exzolt™ Cattle-CA1-treated groups. Exzolt™ Cattle-CA1-treated groups received a single administration at the dose of 2.5 mg/kg. A total of 24 animals were treated with Exzolt™ Cattle-CA1 across these 3 studies. The product demonstrated 100% effectiveness within the first week after Exzolt™ Cattle-CA1 administration. Length of consistent 100% persistent effectiveness ranged from 37 days to approximately 110 days post-treatment.

Out of the 17 studies listed above, 13 field effectiveness studies conducted in Brazil and Australia utilizing natural infestations of *R. microplus* were evaluated. These studies were conducted between 2017 and 2023. In each study, animals were group-housed and randomly assigned to control and Exzolt™ Cattle-CA1-treated groups. Exzolt™ Cattle-CA1-treated groups received a single administration at the dose of 2.5 mg/kg. Approximately 220 animals were treated with Exzolt™ Cattle-CA1 across these 13 studies. The product demonstrated 100% effectiveness within the first week after Exzolt™ Cattle-CA1 administration. Length of consistent 100% persistent effectiveness ranged from 28 days to 70 days post-treatment.

Out of the 17 studies listed above, one rain exposure study was conducted in Brazil in 2021 and utilized induced infestations of *R. microplus* as the clinical endpoint to determine whether rainfall after administration of Exzolt™ Cattle-CA1 had a negative impact on effectiveness. This study is described under the "Effect of Weather on Efficacy" section below.

3. Effect of Weather on Efficacy

A study was conducted to specifically determine the potential effect of rainfall within 24 hours of administration on the efficacy of Exzolt™ Cattle-CA1. In this study, rainfall was simulated at a rate of 10 to 11 mm/10 minutes for 20 minutes. This rate was considered "worst-case" because it is equivalent to the mean maximum hourly rainfall in the United States (based on a U.S. Department of Interior Geological Survey). This study demonstrated that under these conditions, no effect on product efficacy would be expected.

Rain exposure study

Title: Evaluation of Rain Exposure Impact on the Efficacy of a Fluralaner Formulation Administered via Pour On Once, at a Dose of 1 mL/20 kg Body Weight (Corresponding to 2.5 mg Fluralaner/kg), against Artificially Induced *Rhipicephalus (B.) microplus* Infestations on Cattle. (Study No. S19152-00)

Study Dates: May 10, 2021 to August 25, 2021

Study Location: Formiga, Brazil

Study Design:

Objective: To evaluate the impact of rain exposure on the therapeutic and persistent efficacy of topically applied Exzolt™ Cattle-CA1 against artificially induced *R. microplus* tick infestations on beef cattle.

Study Animals: Thirty beef cross-bred bulls, estimated 7 to 16 months of age and weighing between 154 and 385 lbs. (70 to 175 kg) at the start of the study, were enrolled.

Experimental Design: The study was a masked, randomized, negative-controlled study consisting of four Exzolt™ Cattle-CA1-treated groups and one control group. Animals were individually housed for the duration of the study and the experimental unit was the individual animal. Animals were blocked by total number of ticks collected per animal over three days (Days -3, -2, and -1) pre-treatment. Within each block, animals were randomized into one of five treatment groups. There were six blocks of five animals each, as described in Table II.2 below. This study was conducted in accordance with Good Clinical Practice (GCP).

Table II.2. Study Groups

Treatment group	Test Article	Post-Treatment Time of Simulated Rainfall	Number of Animals
A	Exzolt™ Cattle-CA1 (2.5 mg fluralaner/kg body weight)	6 hours (±1 hour)	6
B	Exzolt™ Cattle-CA1 (2.5 mg fluralaner/kg body weight)	12 hours (±1 hour)	6
C	Exzolt™ Cattle-CA1 (2.5 mg fluralaner/kg body weight)	24 hours (±1 hour)	6
D	Exzolt™ Cattle-CA1 (2.5 mg fluralaner/kg body weight)	NA[1]	6
E	Dyed saline control (2.5 mg saline/kg body weight)	NA	6

[1]NA, Not Applicable

Prior to treatment on Day 0, all study animals were infected with approximately 5,000 *R. microplus* larvae ≤8 weeks of age 3 times a week for approximately 5 weeks. Post-treatment, tick infestations were performed twice weekly for approximately 15 weeks.

In groups A, B, and C, rainfall was simulated with animals tied in a stall with water sprayed on them at a rate of 10 to 11 mm/10 minutes for a duration of 20 minutes at the time points indicated in Table II.2. Animals in groups D and E were not exposed to simulated rain. For the duration of the study, animals were housed in a covered barn and not exposed to natural rainfall.

Drug Administration: Exzolt™ Cattle-CA1 (2.5 mg/kg body weight) and saline as control were administered topically once on Day 0 along the dorsal midline from withers to tailhead. The saline was dyed green to maintain masking because Exzolt™ Cattle-CA1 is green. Saline was administered at an equivalent volume as Exzolt™ Cattle-CA1.

Measurements and Observations: General health observations were conducted once daily throughout the study. Treatment site observations were conducted on Day 0 prior to treatment and 5 hours post-treatment, and then on Days 1, 2, 5, 7, 14, 21, and 28. Neurological assessments were performed on Day 0 prior to treatment and 5 hours post-treatment, once daily during the first week after treatment, and then on Days 9, 17, 19, 27, 29, 37, 39, 47, and 49. Daily tick counts were conducted starting on Day 1. Detached, engorged female ticks were counted for each animal.

Statistical Methods: The experimental unit was the individual animal. Counts of detached fully engorged female ticks were analyzed using a linear mixed model

with treatment group as a fixed effect and block as a random effect. The least squares means were used for treatment comparisons. Separate analyses were conducted at each tick count day and for each group treated with Exzolt™ Cattle-CA1. The null hypothesis was that the Exzolt™ Cattle-CA1-treated group and the control group had the same mean tick counts. The hypothesis was tested at α=0.05 (two-sided) significance level.

The percent efficacy on a specific study day starting on Day 7 was calculated using the Henderson-Tilton formula:

Efficacy(%) = $\left[1 - \frac{Ta \times Cb}{Tb \times Ca}\right] \times 100$

Where T_a = Raw mean tick counts of the Exzolt™ Cattle-CA1-treated animals after treatment;

T_b = Raw mean tick counts of the Exzolt™ Cattle-CA1-treated animals prior to treatment;

C_a =Raw mean tick counts of the control animals after treatment;

C_b = Raw mean tick counts of the control animals prior to treatment.

Results: The Exzolt™ Cattle-CA1 treatment groups A, B, C, and D demonstrated efficacies of ≥99% from Day 7 through Day 77 and had mean tick counts significantly different (p≤0.0029) from the control group. A ≥99% threshold for efficacy was used in this study as *R. microplus* is a reportable tick species in the United States.

Adverse Reactions: No adverse reactions were reported in this study.

Conclusions: This study demonstrated that rainfall starting at six hours post-treatment does not impact the therapeutic or persistent efficacy of Exzolt™ Cattle-CA1.

III. TARGET ANIMAL SAFETY

A. Margin of Safety Study

Title: Target Animal Safety Study in Cattle Following Repeated Topical Administration of Fluralaner 50 mg/mL Pour-On Solution for Cattle at Three Different Dose Rates. (Study No. M2001BT)

Study Dates: April 1, 2020 to September 8, 2022

Study Location: Parma, ID

Study Design:

Objective: To evaluate the margin of safety of fluralaner topical solution for cattle administered by topical application 3 times 42 days apart in 6- to 7-month-old beef calves.

Study Animals: Thirty-two healthy beef breed calves (British continental crosses, 16 males, 16 females) approximately 6 to 7 months of age and weighing between 164 and 277 kg (362 to 611 lbs.) at the start of the study were enrolled.

Experimental Design: The study was a masked, randomized margin of safety study with a negative control and was conducted in accordance with the Good Laboratory Practice (GLP) Regulations for Non-Clinical Laboratory Studies. The study animals were individually housed except for four to six hours a day (except on dosing days) when animals of the same treatment group were group-housed to encourage allo-grooming. The experimental unit was the treatment group. The study animals were randomized to the four treatment groups within gender. The four treatment groups were randomly assigned to four location blocks (pen areas A, B, C, or D), and these blocks were randomly assigned to processing order. Within a block, animals were ranked by animal ID without regard to gender and randomized to pen within the pen area (eight pens per pen area). The 1X dose group animals received Exzolt™ Cattle-CA1 at 3.7 mg/kg; 3X dose group animals received Exzolt™ Cattle-CA1 at 11.1 mg/kg; and 5X dose group animals received Exzolt™ Cattle-CA1 at 18.5 mg/kg. The 1X group received 3.7 mg/kg instead of the labeled dose of 2.5 mg/kg because of the potential for rounding up when using the dose bands on the provided dosing chart. The smallest animal in a dosing band could receive up to 3.7 mg/kg. The control group animals received saline at the same dose volume as would be required for a 5X group animal of the same body weight; the saline was dyed green to maintain masking because Exzolt™ Cattle-CA1 is green.

Drug Administration: Test and control articles were administered topically once on Days 0, 42, and 84 along the dorsal midline from withers to tailhead.

Measurements and Observations: General health observations were conducted twice daily from Day -13 to the end of the study; individual body weights were determined on Days -8, -1, 21, 41, 63, 83, and 97/98. Feed and water consumption were recorded daily from the start of acclimation (Day -14) to the end of the study. Physical examinations were conducted on Days -14, -1, 1, 4, 7, 14, 21, 28, 35, 41, 43, 46, 49, 56, 63, 70, 77, 83, 85, 88, 91, and 96. Neurological examinations which included a scoring system for mental status, coordination, menace response, and pupillary light response, as well as evaluation of eye position, eye movement, position of the head and tongue, salivation/drooling or bloat, and symmetry of the face were scheduled on the same days as physical examinations, plus on the days of and during the week after dosing, i.e., Study Days 0 (4 to 6 hours post dosing), 2, 3, 5, 6, 42 (4 to 6 hours post dosing), 44, 45, 47, 48, 84 (4 to 6 hours post dosing), 86, 87, 89, and 90. During acclimation on Study Days -9 and -1, blood samples were collected from individual animals for clinical pathology analysis and for fluralaner bioanalysis. Urine samples for urinalysis and fecal samples for fecal occult blood were collected on Study Days -8 and -1. During the treatment period, blood (including for fluralaner bioanalysis) and fecal samples were collected on Study Days 6, 13, 41, 48, 55, 83, 90, and 96. Urine samples were collected on the same

schedule. Cattle were necropsied on Study Day 97 or 98. A complete gross examination was performed, and a complete set of tissues and any abnormal tissue or gross lesions were collected for histopathologic examination. Weights were taken of brain, heart, kidneys, liver (with drained gall bladder) and spleen, and organ to body weight ratios (%) were calculated. The plasma concentrations of fluralaner were measured using a validated liquid chromatography–mass spectrometry (LCMS/MS) method and the pharmacokinetic parameters were estimated using a non-compartmental analysis.

Statistical Methods: No statistical analysis was conducted because there was only one experimental unit per treatment group. For the data from individual animals, summary results of continuous variables for each treatment group, including the number of animals, mean, standard deviation, median, minimum, and maximum values, were provided by sex; summary results of continuous variables measured repeatedly for each treatment group were provided by sex and by time; frequency summaries for categorical variables for each treatment group were provided by sex and by time if applicable.

Results:

Clinical observations: There were no clinically relevant treatment-related effects on physical examination parameters, neurological examination parameters, food/water consumption, body weight, clinical pathology parameters, urinalysis, or fecal occult blood. Abnormal general health findings in individual animals included ruminal tympany, fever, hematochezia, and pneumonia but these were sporadic across all treatment groups and considered typical findings in this age and class of animal and not associated with the test article. There were treatment-related skin abnormalities at the test article application site. These reactions were dose dependent and in the 1X animals were not seen until after the second dose. Reactions were limited mostly to skin scurfing and scabbing with some skin thickening and loss of elasticity. No skin lesions required treatment. Because animals were necropsied two weeks after the last treatment, most skin lesions had not completely resolved by the end of the study.

Conclusions: The study demonstrates that Exzolt™ Cattle-CA1 is safe for use in beef cattle when administered topically at the recommended label dose with an acceptable margin of safety.

B. Reproductive Safety Study

Title: Reproductive Safety Study in Female Cattle Following Topical Administration of Fluralaner 50 mg/mL Pour-On Solution for Cattle. (Study No. M2001BP)

Study Dates: November 5, 2020 to November 12, 2021

Study Location: Parma, ID

Study Design:

Objective: To evaluate the reproductive safety of fluralaner topical solution for cattle administered by topical application at 0X or 3X the maximum anticipated dose to

female cattle during breeding, early- to mid-first trimester, or third trimester of pregnancy.

Study Animals: Two-hundred healthy beef breed cows (British continental crosses) 3 to 11 years old and weighing between 431.5 and 723.0 kg (951.3 to 1593.9 lbs.) at the start of the study were enrolled.

Experimental Design: The study was a masked, randomized reproductive safety study with a negative control and was conducted in accordance with the Good Laboratory Practice (GLP) Regulations for Non-Clinical Laboratory Studies. The study animals were housed by treatment group and cohort. Calves were housed with the dams. The study was split into 2 cohorts of 100 animals each, with 20 animals per treatment group in each cohort. The study animals were randomized in pairs (for cohort 1 and cohort 2) to the five treatment groups.

Table III.1. Treatment Groups (Day 0 Is Day of Artificial Insemination)

Group/Dose/Animal #	Dose Study Day -13±1	Dose Study Day 11±1	Dose Study Day 38±1	Dose Study Day 218±1
T01; 0X (control)/40	Saline	Saline	Saline	Saline
T02; 3X/40	Fluralaner topical solution	Saline	Saline	Saline
T03; 3X/40	Saline	Fluralaner topical solution	Saline	Saline
T04; 3X/40	Saline	Saline	Fluralaner topical solution	Saline
T05; 3X/40	Saline	Saline	Saline	Fluralaner topical solution

Study Day 0 was the day of artificial insemination. The animals in the test article groups received fluralaner topical solution at 11.1 mg/kg (3X the maximum labeled dose) once either during estrus (Day -13±1), early in first trimester (Day 11±1), during mid-first trimester (Day 38±1), or in the third trimester of pregnancy (Day 218±1). The control group animals received saline at the same dose volume as the 3X group. At each dosing day, saline was also administered to cattle of test article groups which were not scheduled for treatment with test article on those days. The saline was dyed green to maintain masking because fluralaner topical solution is green.

Drug Administration: Test and control articles were administered topically along the dorsal midline from withers to tailhead.

Measurements and Observations: General health observations were conducted twice daily on cows throughout the entire study. Conception rate, pregnancy rate, abortion rate, stillbirth rate, dystocia scores, and calf health and body weight at birth and 30 days post-partum were evaluated. Three scheduled pregnancy checks (Day 50±2, Day 91±2, and Day 216±2) were performed.

Statistical Methods: The experimental unit was the treatment group by cohort. No inferential statistical analyses were conducted because there were only two experimental units per treatment group. For cow- and calf-related data, summary statistics including number of observations, median, mean, standard deviation, maximum, and minimum for data of continuous variable and frequency distributions for categorical data were calculated by treatment group and over time, where applicable.

Results:

Table III.2. Results of Reproductive Variables (Day 0 Is Day of Artificial Insemination)

Variable	T01 (Control)	T02 (Treated Day -13±1)	T03 (Treated Day 11±1)	T04 (Treated Day 38±1)	T05 (Treated Day 218±1)
Conception Rate (# Pregnant Day 50±2 / # Bred)[1]	30/40 75%	31/40 77.5%	34/40 85%	27/40 67.5%	N/A
Abortion Rate (# Lost Pregnancies / # Pregnant on Day 50±2)[2]	0/30 0%	2/31 6.5%	2/34 5.9%	2/27 7.4%	0/30 0%
Stillbirth Rate (# Full-Term Deliveries with Stillborn Calves / # Pregnant Day 50±2)[2]	3/30 10%	0/31 0%	0/34 0%	1/27 3.7%	0/30 0%
Calving Rate (# Live Full Term Calves Delivered / # Pregnant Day 50±2)[2]	27/30 90%	29/31 93.5%	32/34 94.1%	24/27 88.9%	30/30 100%
Calf Birth Weights (Mean)	32.76 kg	33.87 kg	32.55 kg	32.39 kg	33.66 kg
Calf Weights at 30 Days (Mean)	67.63 kg	66.84 kg	64.59 kg	67.42 kg	66.15 kg

[1]Conception Rate for Groups T01, T02, T03, and T04 only
[2]Rates for T05 calculated using denominator as "# Pregnant on Day 218" due to timing of test article administration

Clinical observations: There were no clinically relevant treatment-related effects on the reproductive variables tabulated above. There were also no clinically significant differences in dystocia scores between treated groups and the control group. While abortions did occur in the test article treatment groups, the rates were low, typical for the source herd, and occurred typically more than two months after test article administration. Abnormal general health findings in cows included singular cases of vaginal cyst, vaginal prolapse, and nasal squamous cell carcinoma. Three calves had hematochezia with or without diarrhea. One calf in treatment group 3 was found dead prior to the 24-hour post-partum examination after a normal birth. At necropsy, the calf was found with an empty abomasum and it was concluded this calf died from failure to nurse. These adverse events were considered typical findings for cows and neonatal calves and not considered related to the test article.

Conclusions: The study demonstrates that Exzolt™ Cattle-CA1 is safe for use in reproducing female beef cattle when administered topically at the labeled dose.

IV. HUMAN FOOD SAFETY

A. Microbial Food Safety

This drug is not an antimicrobial. Therefore, the Agency determined that a microbial food safety assessment was not required.

B. Toxicology

Reassessment of the acceptable daily intake (ADI) was not needed for this approval. The codified ADI for total residue of fluralaner is 10 micrograms per kilogram of body weight per day (µg/kg bw/day).

The sponsor requested revision of the safe concentrations for total residues of fluralaner in the edible tissues using a new partition of the ADI. The calculation of the tissue safe concentrations is based on the "General Principles for Evaluating the Human Food Safety of New Animal Drugs Used in Food-Producing Animals" (FDA/CVM, Guidance for Industry #3, May 2022) and reflects the partition (meat, milk, and eggs) requested by the drug sponsor. The safe concentration for total residues of fluralaner in parts per billion (ppb) in each edible tissue of beef cattle is calculated using the following formula:

$$\text{Safe Concentration} = \frac{\text{Percent Partition} \times \text{ADI x Human Body Weight}}{\text{Food Consumption Value}}$$

The safe concentrations for total residues of fluralaner in the individual edible tissues of beef cattle are 440 ppb for muscle, 1,320 ppb for liver, 2,640 ppb for kidney, and 2,640 ppb for fat. These values reflect the partition of the ADI between meat (22% of the ADI) and eggs (71% of the ADI) and the amount reserved for milk (7% of the ADI).

The FOI Summary for the original approval of NADA 141-607, dated July 17, 2025, contains summaries of all toxicology studies and information.

C. Residue Chemistry

1. Summary of Residue Chemistry Studies

   a. Total Residue and Metabolism Studies

   Title: A Pivotal Metabolism Study in Beef Cattle Following a Single Topical Application of 5.6 mg [^{14}C]-Fluralaner/kg Body Weight.
   (Study No. S17157-00-CFP-MET-RM)

   Study Dates: April 15, 2020 to July 29, 2025

   Study Location: Tranent, East Lothian, United Kingdom

   Study Design:

   Objective: The objective of this study was to determine concentrations of total residues of fluralaner and characterize fluralaner metabolites in cattle muscle, liver, fat, and kidney tissues following topical administration of fluralaner at a dose of 5.6 mg fluralaner/kg body weight applied topically in a narrow strip along the dorsal midline from the withers to the base of the tail.

   Study Animals: Twenty-eight head of cattle (14 males and 14 females) were used in this study. Cattle were various cross breeds and weighed 279.86 ± 23.40 kg 1 day prior to dosing.

   Experimental Design: The study was conducted in accordance with Good Laboratory Practices (GLPs; 21 CFR § 58). Cattle were assigned to one of six treatment groups (Table IV.1).

   Table IV.1. Experimental Groups

Group	Number of Cattle	Slaughter Time
1	2 males and 2 females	7 days after dose
2	2 males and 2 females	14 days after dose
3	2 males and 2 females	21 days after dose
4	2 males and 2 females	35 days after dose
5	2 males and 2 females	42 days after dose
6	2 males and 2 females	60 days after dose

   Drug Administration: Cattle were dosed topically with a solution containing a mixture of [^{14}C]-fluralaner and unlabeled fluralaner on a single occasion. The dosing solution was applied topically on the backline between the withers and base of the tail at a dose of 5.6 mg fluralaner/kg body weight.

   Measurements and Observations: At the assigned withdrawal period (Table IV.1), cattle were slaughtered by captive bolt followed by pithing and exsanguination. Samples of muscle, liver, fat, and kidney were collected from each animal, including tissue from three areas along the application site (A, B and C).

   Concentrations of total residues of fluralaner were determined by combustion followed by liquid scintillation counting.

Tissue samples were extracted and subjected to metabolite profiling by high-pressure liquid chromatography coupled with a radioactivity detector (radio-HPLC).

Parent fluralaner and four major fluralaner metabolites were isolated from tissue samples by liquid chromatography and structurally elucidated by high resolution mass spectrometry.

Concentrations of parent fluralaner were determined in samples by liquid chromatography with mass spectrometry detection (LC-MS/MS) using a validated analytical procedure.

Results: Total residues of fluralaner (Tables IV.2 and IV.3) and parent fluralaner (Tables IV.4 and IV.5) were detected in tissue samples and depleted over time. These data enabled the relationship between parent fluralaner and total residues of fluralaner to be established (Tables IV.6 and IV.7). The ratio between parent fluralaner (marker residue) and total residues of fluralaner (M:T) ranged between 0.69 and 1 for muscle, 0.30 and 0.66 for liver, 0.66 and 1 for fat, and 0.62 and 0.82 for kidney.

Table IV.2. Mean (± standard deviation) Concentrations (ppb) of Total Residues of Fluralaner in Tissues from Cattle Dosed Topically with a Mixture of [^{14}C]-fluralaner and Unlabeled Fluralaner on the Backline between the Withers and Head of the Tail at a Rate of 5.6 mg Fluralaner/kg Body Weight

WDP[1] (days)	Remote Muscle	Liver	Renal Fat	Mesenteric Fat	Subcutaneous Fat	Kidney
7	453.97 ± 221.58 n = 4	4410.6 ± 2235.66 n = 4	3573.99 ± 1342.05 n = 4	2532.52 ± 582.39 n = 4	826.3 ± 492.79 n = 4	1728.09 ± 859.58 n = 4
14	192.8 ± 44.9 n = 4	2287.64 ± 488.13 n = 4	1562.45 ± 330.97 n = 4	1131.7 ± 234.12 n = 4	670.19 ± 299.63 n = 4	717.01 ± 67.69 n = 4
21	126.25 ± 46.4 n = 4	1554.04 ± 560.19 n = 4	1160.27 ± 384.82 n = 4	904.15 ± 315.87 n = 4	521.52 ± 456.59 n = 4	504.61 ± 181.92 n = 4
35	25.12 ± 14.31 n = 4	318.91 ± 116.2 n = 4	206.2 ± 137.19 n = 4	165.18 ± 127.02 n = 4	130.4 ± 123.92 n = 4	90.79 ± 55.28 n = 4
42	26.12 ± 13.28 n = 4	335.98 ± 143.3 n = 4	219.15 ± 124.14 n = 4	188.83 ± 114.7 n = 4	75.08 ± 93.22 n = 4	96.78 ± 53.95 n = 4
60	3.4 ± 2.6 n = 4	48.22 ± 42.52 n = 4	17.79 ± 18.1 n = 4	10.03 ± 3.44 n = 4	12.42 ± 4.74 n = 4	10.28 ± 8.52 n = 4

[1]WDP, Withdrawal period

Table IV.3. Mean (± standard deviation) Concentrations (ppb) of Total Residues of Fluralaner in Application Site Tissues from Cattle Dosed Topically with a Mixture of [^{14}C]-fluralaner and Unlabeled Fluralaner on the Backline between the Withers and Head of the Tail at a Rate of 5.6 mg Fluralaner/kg Body Weight

WDP[1] (days)	APS[2] A Muscle	APS B Muscle	APS C Muscle	Composite APS Fat	APS A Fat	APS B Fat	APS C Fat
7	471.57 ± 242.45 n = 4	394.68 ± 187.16 n = 4	447.5 ± 216.7 n = 4	1224.27 ± 719.75 n = 4	NC[3]	NC	NC
14	222.17 ± 41.14 n = 4	166.77 ± 38.22 n = 4	223.54 ± 47.04 n = 4	592.37 ± 217.38 n = 4	NC	NC	NC
21	91.61 ± 22.19 n = 4	109.61 ± 38.57 n = 4	156.4 ± 51.25 n = 4	350.24 ± 134.75 n = 3	1024.66 n = 1	797.27 n = 1	835.495 n = 1
35	35.68 ± 25.27 n = 4	22.12 ± 14.8 n = 4	26.35 ± 13.27 n = 4	129.24 ± 113.15 n = 4	NC	NC	NC
42	30.7 ± 15.49 n = 4	22.86 ± 12.76 n = 4	34.61 ± 20.57 n = 4	124.85 ± 91.34 n = 2	73.02 ± 54.55 n = 2	62.27 ± 40.58 n = 2	89.55 ± 28.04 n = 2
60	3.68 ± 3.03 n = 4	3.11 ± 2.02 n = 4	3.71 ± 3.02 n = 4	5.39 ± 0.74 n = 3	20.75 n = 1	26.72 n = 1	20.08 n = 1

[1]WDP, Withdrawal period
[2]APS, Application site
[3]NC, Not collected. Composited sample collected instead

Table IV.4. Mean (± standard deviation) Concentrations (ppb) of Parent Fluralaner in Tissues from Cattle Dosed Topically with a Mixture of [^{14}C]-fluralaner and Unlabeled Fluralaner on the Backline between the Withers and Head of the Tail at a Rate of 5.6 mg Fluralaner/kg Body Weight

WDP[1] (days)	Remote Muscle	Liver	Renal Fat	Mesenteric Fat	Subcutaneous Fat	Kidney
7	375.4 ± 189.66 n = 4	2404.7 ± 1845.55 n = 4	3661.25 ± 1764.67 n = 4	2250.25 ± 573.06 n = 4	786.72 ± 637.38 n = 4	1301.28 ± 674.87 n = 4
14	150.85 ± 38.35 n = 4	1196.05 ± 229.39 n = 4	1468.5 ± 289.58 n = 4	994.45 ± 169.52 n = 4	601.6 ± 284.44 n = 4	520.08 ± 33.64 n = 4
21	100.88 ± 40.83 n = 4	773.4 ± 245.48 n = 4	1019.5 ± 334.6 n = 4	895.52 ± 333.38 n = 4	431.92 ± 301.93 n = 4	351.4 ± 136.76 n = 4
35	25.06 ± 8.28 n = 3	171.47 ± 45.21 n = 3	243.23 ± 100.96 n = 3	189.24 ± 111.77 n = 3	162.2 ± 62.51 n = 2	83.73 ± 28.76 n = 3
42	26.83 ± 3.46 n = 2	175.16 ± 78.7 n = 3	201.19 ± 123.66 n = 4	162.8 ± 98.17 n = 4	133.6 n = 1	81.31 ± 35.02 n = 3
60	<LOQ[2]	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ

[1]WDP, Withdrawal period
[2]LOQ, less than the limit of quantification (Muscle LOQ = 8.49 ppb; Liver LOQ = 43.1 ppb; Fat LOQ = 40.3 ppb; Kidney LOQ = 24.0 ppb)

Table IV.5. Mean (± standard deviation) Concentrations (ppb) of Parent Fluralaner in Application Site Tissues from Cattle Dosed Topically with a Mixture of [^{14}C]-fluralaner and Unlabeled Fluralaner on the Backline between the Withers and Head of the Tail at a Rate of 5.6 mg Fluralaner/kg Body Weight

WDP[1] (days)	APS[2] A Muscle	APS B Muscle	APS C Muscle	Composite APS Fat	APS A Fat	APS B Fat	APS C Fat
7	405.05 ± 208.09 n = 4	330.7 ± 176.69 n = 4	358.05 ± 174.83 n = 4	993.6 ± 607.01 n = 4	NC[3]	NC	NC
14	168.43 ± 29.45 n = 4	131.78 ± 26.45 n = 4	175.35 ± 38.96 n = 4	461.65 ± 152.3 n = 4	NC	NC	NC
21	128.03 ± 47.69 n = 4	92.44 ± 33.36 n = 4	128.36 ± 44.12 n = 4	342.47 ± 88.87 n = 3	712.30 n = 1	984.00 n = 1	689.90 n = 1
35	32.72 ± 14.71 n = 3	26.58 ± 9.87 n = 2	24.36 ± 7.19 n = 3	186.65 ± 95.11 n = 2	NC	NC	NC
42	25.5 ± 8.82 n = 3	23.62 n = 1	28.58 ± 13.52 n = 3	142.6 n = 1	96.69 n = 1	71.64 n = 1	91.11 n = 1
60	<LOQ[4]	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ

[1]WDP, Withdrawal period
[2]APS, Application site
[3]NC, Not collected. Composited sample collected instead.; [4]<LOQ, less than the limit of quantification (Muscle LOQ = 8.49 ppb; Fat LOQ = 40.3 ppb; Kidney LOQ = 24.0 ppb)

Table IV.6. Ratio (mean ± standard deviation and range) between Parent Fluralaner (the marker residue) and Total Residues of Fluralaner in Tissues from Cattle Dosed Topically with a Mixture of [^{14}C]-fluralaner and Unlabeled Fluralaner on the Backline between the Withers and Head of the Tail at a Rate of 5.6 mg Fluralaner/kg Body Weight

WDP[1] (days)	Remote Muscle	Liver	Renal Fat	Mesenteric Fat	Subcutaneous Fat	Kidney
7	0.82 ± 0.02 0.8 - 0.84	0.51 ± 0.16 0.3 - 0.66	1.00 ± 0.1 0.90 - 1	0.89 ± 0.06 0.83 - 0.96	0.87 ± 0.19 0.66 - 1	0.75 ± 0.04 0.69 - 0.78
14	0.78 ± 0.03 0.73 - 0.81	0.53 ± 0.03 0.49 - 0.57	0.94 ± 0.04 0.89 - 0.97	0.88 ± 0.09 0.82 - 1	0.90 ± 0.06 0.82 - 0.96	0.73 ± 0.03 0.70 - 0.75
21	0.79 ± 0.07 0.69 - 0.84	0.5 ± 0.03 0.45 - 0.53	0.88 ± 0.08 0.77 - 0.96	1 ± 0.12 0.87 - 1	0.91 ± 0.16 0.71 - 1	0.69 ± 0.05 0.62 - 0.73
35	0.79 ± 0.09 0.7 - 0.88	0.45 ± 0.1 0.35 - 0.55	0.94 ± 0.08 0.86 - 1	0.90 ± 0.06 0.83 - 0.94	0.71 ± 0.01 0.70 - 0.72	0.74 ± 0.07 0.68 - 0.82
42	0.72 ± 0.01 0.72 - 0.73	0.45 ± 0.05 0.39 - 0.49	0.89 ± 0.1 0.81 - 1	0.87 ± 0.07 0.78 - 0.93	0.63	0.71 ± 0.01 0.70 - 0.72
60	NA[2]	NA	NA	NA	NA	NA

[1]WDP, Withdrawal period
[2]NA, Not applicable because marker residue was not quantifiable

Table IV.7. Ratio (mean ± standard deviation and range) between Parent Fluralaner (the marker residue) and Total Residues of Fluralaner in Application Site Tissues from Cattle Dosed Topically with a Mixture of [^{14}C]-fluralaner and Unlabeled Fluralaner on the Backline between the Withers and Head of the Tail at a Rate of 5.6 mg Fluralaner/kg Body Weight

WDP[1] (days)	APS[2] A Muscle	APS B Muscle	APS C Muscle	Composite APS Fat	APS A Fat	APS B Fat	APS C Fat
7	0.86 ± 0.02 0.83 - 0.88	0.82 ± 0.05 0.77 - 0.88	0.8 ± 0.01 0.79 - 0.81	0.80 ± 0.09 0.66 - 0.86	NC[4]	NC	NC
14	0.76 ± 0.06 0.70 - 0.84	0.79 ± 0.05 0.75 - 0.85	0.78 ± 0.02 0.76 - 0.80	0.79 ± 0.05 0.72 - 0.83	NC	NC	NC
21	1.55 ± 0.87 0.78 - 1	0.84 ± 0.02 0.82 - 0.86	0.82 ± 0.05 0.78 - 0.89	1.07 ± 0.5 0.75 - 1	0.70	1.23	0.83
35	0.72 ± 0.05 0.69 - 0.77	0.81 ± 0.02 0.79 - 0.83	0.74 ± 0.11 0.61 - 0.81	0.87 ± 0.03 0.84 - 0.89	NC	NC	NC
42	0.7 ± 0.01 0.70 - 0.71	0.77	0.68 ± 0.06 0.64 - 0.76	0.75	0.87	0.79	0.83
60	NA[3]	NA	NA	NA	NA	NA	NA

[1]WDP, Withdrawal period
[2]APS, Application site
[3]NA, Not applicable because marker residue was not quantifiable
[4]NC, Not collected. Composited sample collected instead

Parent fluralaner and four major metabolites were detected in edible cattle tissues. Mass spectrometry indicated that the major metabolites are the following: Metabolite 4 (hydroxy-fluralaner), AH362502 (monohydroxy-fluralaner), Metabolite 12 (glutathione conjugate of fluralaner), and Metabolite 15 (isoxazole ring-opened fluralaner).

Conclusions: Liver was the tissue in which total residues of fluralaner last depleted to less than their safe concentration. This indicates that, when liver total residue concentrations are less than the liver safe concentration, total residues of fluralaner in the other edible tissues are less than their respective safe concentration. The M:T ratio in meat tissues is variable for fluralaner. When concentrations of total residues of fluralaner in liver and muscle were less than their respective safe concentrations, concentrations of parent fluralaner were approximately 500 ppb and 350 ppb, respectively. These concentrations equate to M:T ratios of approximately 0.38 and 0.80 for liver and muscle, respectively. To assess the safety of application site fat tissues, the concentration of parent fluralaner was determined when the corresponding concentration of total residues of fluralaner in application site fat tissues were less than the fat safe concentration. This concentration of parent fluralaner was determined to be 1800 ppb.

Four major metabolites of fluralaner were detected in edible cattle tissues: Metabolite 4 (hydroxy-fluralaner), AH362502 (monohydroxy-fluralaner), Metabolite 12 (glutathione conjugate of fluralaner), and Metabolite 15 (isoxazole ring-opened fluralaner).

The data from this study indicate liver and parent fluralaner are an appropriate target tissue and marker residue, respectively, for monitoring the human food safety of edible meat tissues obtained from cattle treated with fluralaner topical solution. The data also indicate that a liver tolerance of 500 ppb and a muscle tolerance of 350 ppb are protective of human health.

b. Comparative Metabolism Study

Title: [^{14}C]-Fluralaner: The Biliary Elimination of Radioactivity in the Beagle Dog Following Multiple Oral Administrations of [^{14}C]-Fluralaner. (Study No. S22162-00)

Study Dates: December 18, 2023 to July 25, 2025

Study Location: Tranent, East Lothian, United Kingdom

Study Design:

Objective: The objective of this study was to confirm that the toxicological animal species used to establish the ADI (i.e., dogs) was exposed to the major metabolites detected in edible cattle meat tissues (i.e., autoexposure).

Study Animals: One male beagle dog (10.03 kg) was used in this study.

Experimental Design: The study was conducted in accordance with GLPs (21 CFR § 58).

Drug Administration: The dog was administered [^{14}C]-fluralaner in gelatin capsules at a dose of 25 mg fluralaner/kg body weight once daily for seven consecutive days (Study Days 1-7).

Measurements and Observations: Bile duct cannulation surgery was performed on Study Day 14. Bile samples were collected at 24, 48, 72, and 96 hours after the last dose. On Study Day 11, the dog was euthanized by an overdose of pentobarbital.

Samples were extracted and subjected to radio-HPLC. Metabolites corresponding to the major metabolites detected in cattle tissues were structurally elucidated by high resolution mass spectrometry.

Results: The major metabolites detected in cattle tissues (Metabolite 4 (hydroxy-fluralaner), AH362502 (monohydroxy-fluralaner), Metabolite 12 (glutathione conjugate of fluralaner), and Metabolite 15 (isoxazole ring-opened fluralaner)) were detected by high resolution mass spectrometry in bile samples collected from the dog administered fluralaner.

Conclusions: Dogs orally administered fluralaner are exposed to the fluralaner metabolites that humans can be exposed to through the consumption of meat produced by cattle treated with fluralaner. Therefore, the ADI determined from data generated in dogs is appropriate for assessing the human food safety of meat from cattle treated with fluralaner.

c. Studies to Establish the Withdrawal Period

(1) Tissue Residue Depletion Study

Title: Residue Depletion Study of Fluralaner in Cattle Tissues Following Topical Administration of Fluralaner Topical Solution.
(Study No. S19115-00)

Study Dates: December 11, 2020 to January 19, 2023

Study Location:

In-life phase: Parma, ID
Tissue analysis: Rahway, NJ

Study Design:

Objective: The objective of this study was to determine parent fluralaner concentrations in cattle tissues following topical administration of fluralaner at a dose of up to 3.7 mg fluralaner/kg body weight applied topically in a narrow strip along the dorsal midline from the withers to the base of the tail during cold-weather conditions.

Study Animals: Sixty-eight Angus cattle (34 steers and 34 heifers) were used in this study. Cattle weighed 330.57 ± 19.37 kg on Day -2 (Day 0 = dosing).

Experimental Design: The study was conducted in accordance with GLPs (21 CFR § 58). Cattle were assigned to a control untreated group (Group 1) or 1 of 11 treatment groups (Groups 2-12; Table IV.8).

Table IV.8. Experimental Groups for Cattle Dosed Topically with Fluralaner at a Dose of up to 3.7 mg Fluralaner/kg Body Weight

Group	Number of Cattle	Slaughter Time
1 (control)	1 steer and 1 heifer	Study Day 1
2	3 steers and 3 heifers	7 days after dose
3	3 steers and 3 heifers	14 days after dose
4	3 steers and 3 heifers	21 days after dose
5	3 steers and 3 heifers	35 days after dose
6	3 steers and 3 heifers	42 days after dose
7	3 steers and 3 heifers	61 days after dose
8	3 steers and 3 heifers	79 days after dose
9	3 steers and 3 heifers	112 days after dose
10	3 steers and 3 heifers	133 days after dose
11	3 steers and 3 heifers	154 days after dose
12	3 steers and 3 heifers	182 days after dose

Drug Administration: Cattle were treated with a nominal dose of 4 mg fluralaner/kg body weight. The dosing solution (50 mg fluralaner/mL) was administered topically on the backline between the withers and base of the tail using a single-use syringe. The average dose administered was 4.10±0.06 mg fluralaner/kg body weight.

Measurements and Observations: At the assigned withdrawal period (Table IV.8), cattle were slaughtered by captive bolt followed by exsanguination. Samples of muscle, liver, renal fat, mesenteric fat, kidney, heart, and tongue were collected from each animal. Additional muscle and fat samples were obtained from the anterior, middle, and posterior regions of the application site. Concentrations of parent fluralaner were determined in individual samples by LC-MS/MS using validated analytical procedures.

Statistical Methods: Regression analysis was performed on liver, muscle, and application site fat data separately. Then, the upper tolerance limit for the 99th percentile with 95% confidence (99/95 UTL) was calculated from each of the regression models generated for liver, muscle, and application site fat.

Results: Parent fluralaner was detected in the samples collected and depleted as the withdrawal period progressed (Table IV.9 and Table IV.10). The muscle and liver 99/95 UTLs were less than the muscle and liver tolerances determined from Study S17157-00, respectively, at 20 and 98 days after administration, respectively. The application site muscle UTLs were less than the muscle tolerance determined from Study S17157-00 at 30 days after administration. The application site fat UTLs were less than the previously determined safety number (1,800 ppb) at 38 days after administration.

Table IV.9. Mean (± standard deviation) Concentrations (ppb) of Parent Fluralaner in Non-Application Site Tissues from Cattle Dosed Topically with Fluralaner at a Dose of up to 3.7 mg Fluralaner/kg Body Weight

WDP[1] (days)	Remote Muscle	Liver	Renal Fat	Mesenteric Fat	Kidney	Heart	Tongue
7	44.62 ± 17.63 n = 5	329.80 ± 179.85 n = 6	285.38 ± 184.82 n = 6	319.13 ± 201.78 n = 6	134.78 ± 79.59 n = 6	86.42 ± 51.4 n = 6	110.55 ± 76.15 n = 6
14	38.4 ± 23.27 n = 6	351.63 ± 220.15 n = 6	261.2 ± 160.27 n = 6	316.65 ± 209.66 n = 6	143.22 ± 89.82 n = 6	90.6 ± 54.26 n = 6	105.58 ± 60.22 n = 6
21	90.62 ± 153.58 n = 6	788.07 ± 1293.03 n = 6	571.45 ± 897.64 n = 6	780.85 ± 1332.83 n = 6	303.77 ± 481.22 n = 6	190.5 ± 308.17 n = 6	239.13 ± 376.76 n = 6
35	16.22 ± 4.25 n = 5	128.02 ± 42.51 n = 6	106.65 ± 40.53 n = 6	131.62 ± 53.5 n = 6	51.92 ± 19.23 n = 6	33.3 ± 12.72 n = 6	39.35 ± 16.29 n = 6
42	37.95 ± 24.18 n = 4	190.70 ± 226.01 n = 6	192.05 ± 109.57 n = 4	324 ± 221.86 n = 4	126.25 ± 71.07 n = 4	93.38 ± 54.31 n = 4	77.57 ± 60.36 n = 5
61	10.04 ± 0.47 n = 3	55.50 ± 36.71 n = 6	54.8 ± 17.68 n = 2	80.42 ± 17.27 n = 4	32.13 ± 1.97 n = 3	20.52 ± 3.12 n = 4	24.35 ± 7.94 n = 4
79	15 n = 1	36.94 ± 53.76 n = 6	77.3 ± 50.49 n = 2	84.1 ± 42.28 n = 2	54.5 n = 1	24.25 ± 14.5 n = 2	27.2 ± 18.24 n = 2
112	<LOQ[2]	28.52 ± 31.22 n = 4	63.9 n = 1	71.7 n = 1	30.6 n = 1	19.4 n = 1	16.18 ± 10.63 n = 2
133	<LOQ	6.28 n = 1	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
154	<LOQ	4.32 ± 1.46 n = 3	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
182	<LOQ	9.45 ± 6.12 n = 4	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ

[1]WDP, Withdrawal period

[2]<LOQ, less than the limit of quantification (Muscle LOQ = 8.49 ppb; Liver LOQ = 2.49 ppb; Fat LOQ = 40.3 ppb; Kidney LOQ = 24.0 ppb)

Table IV.10. Mean (± standard deviation) Concentrations (ppb) of Parent Fluralaner in Application Site Tissues from Cattle Dosed Topically with Fluralaner at a Dose of up to 3.7 mg Fluralaner/kg Body Weight

WDP[1] (days)	APS[2] Muscle Anterior	APS Muscle Middle	APS Muscle Posterior	APS Fat Anterior	APS Fat Middle	APS Fat Posterior
7	43.36 ± 18.45 n = 5	41.72 ± 17.35 n = 5	41.22 ± 17.1 n = 5	276.6 ± 172.62 n = 6	280.42 ± 170.24 n = 6	280 ± 168.79 n = 6
14	42.3 ± 22.12 n = 5	42.64 ± 22.43 n = 5	42.4 ± 20.61 n = 5	278.33 ± 149.7 n = 6	263.35 ± 173.32 n = 6	271.32 ± 180.45 n = 6
21	84.83 ± 139.69 n = 6	83.25 ± 139.2 n = 6	84.82 ± 140.7 n = 6	667.03 ± 1107.93 n = 6	673.28 ± 1125.99 n = 6	701.43 ± 1189.92 n = 6
35	17.3 ± 3.45 n = 5	16.4 ± 3.31 n = 5	16.52 ± 3.64 n = 5	121.27 ± 51.48 n = 6	122.53 ± 55.14 n = 6	119.83 ± 48.28 n = 6
42	41.62 ± 29.42 n = 4	36.8 ± 24.15 n = 4	38.17 ± 27.22 n = 4	277.75 ± 147.45 n = 4	285 ± 160.08 n = 4	288.25 ± 178.25 n = 4
61	11 ± 0.89 n = 3	10.27 ± 1.13 n = 3	10.87 ± 0.99 n = 3	79.25 ± 18.98 n = 4	77.28 ± 14.4 n = 4	76.72 ± 17.05 n = 4
79	13.4 n = 1	14.2 n = 1	15.8 n = 1	87.95 ± 41.08 n = 2	90.25 ± 47.73 n = 2	89.55 ± 45.89 n = 2
112	11.3 n = 1	8.98 n = 1	9.43 n = 1	75 n = 1	74 n = 1	74.1 n = 1
133	<LOQ[3]	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
154	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
182	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ

[1]WDP, Withdrawal period
[2]APS, Application site
[3]<LOQ, less than the limit of quantification (Muscle LOQ = 8.49 ppb; Fat LOQ = 40.3 ppb)

Conclusions: The data support a presumptive 98-day withdrawal period for cattle treated with fluralaner topical solution at a dose of up to 3.7 mg fluralaner/kg body weight applied topically in a narrow strip along the dorsal midline from the withers to the base of the tail.

(2) Tissue Residue Depletion Study

Title: Residue Depletion Study of Fluralaner in Cattle Tissues Following Topical Administration of Fluralaner Topical Solution.
(Study No. S20167-00)

Study Dates: May 7, 2021 to November 21, 2022

Study Location:

In-life phase: Tulare, CA
Tissue analysis: Rahway, NJ

Study Design:

Objective: The objective of this study was to determine parent fluralaner concentrations in cattle tissues following topical administration of fluralaner at a dose of up to 3.7 mg fluralaner/kg body weight applied

topically in a narrow strip along the dorsal midline from the withers to the base of the tail during warm-weather conditions.

Study Animals: Sixty-eight English breed cattle (34 steers and 34 heifers) were used in this study. Cattle weighed 304.41±24.71 kg on Day -2 (Day 0 = dosing).

Experimental Design: The study was conducted in accordance with GLPs (21 CFR § 58). Cattle were assigned to a control untreated group (Group 1) or 1 of 11 treatment groups (Groups 2-12) (Table IV.11).

Table IV.11. Experimental Groups for Cattle Dosed Topically with Fluralaner at a Dose of up to 3.7 mg Fluralaner/kg Body Weight

Group	Number of Cattle	Slaughter Time
1 (control)	1 steer and 1 heifer	Study Day 1
2	3 steers and 3 heifers	7 days after dose
3	3 steers and 3 heifers	14 days after dose
4	3 steers and 3 heifers	21 days after dose
5	3 steers and 3 heifers	35 days after dose
6	3 steers and 3 heifers	42 days after dose
7	3 steers and 3 heifers	61 days after dose
8	3 steers and 3 heifers	79 days after dose
9	3 steers and 3 heifers	112 days after dose
10	3 steers and 3 heifers	133 days after dose
11	3 steers and 3 heifers	154 days after dose
12	3 steers and 3 heifers	182 days after dose

Drug Administration: Cattle were treated with a nominal dose of 4 mg fluralaner/kg body weight. The dosing solution (50 mg fluralaner/mL) was administered topically on the backline between the withers and base of the tail using a single-use syringe. The average dose administered was 4.17±0.09 mg fluralaner/kg body weight.

Measurements and Observations: At the assigned withdrawal period (Table IV.11), cattle were slaughtered by captive bolt followed by exsanguination. Samples of muscle, liver, renal fat, mesenteric fat, kidney, heart, and tongue were collected from each animal. Additional muscle and fat samples were obtained from the anterior, middle, and posterior regions of the application site. Concentrations of parent fluralaner were determined in individual samples by LC-MS/MS using validated analytical procedures.

Statistical Methods: Regression analysis was performed on liver, muscle, and application site fat data separately. Then, the upper tolerance limit for the 99th percentile with 95% confidence (99/95 UTL) was calculated from each of the regression models generated for liver, muscle, and application site fat.

Results: Parent fluralaner was detected in the samples collected and depleted as the withdrawal period progressed (Table IV.12 and Table IV.13). The muscle and liver 99/95 UTLs were less than the muscle and liver tolerances determined from Study S17157-00, respectively, at 17 and 44 days after administration, respectively. The application site muscle UTLs were less than the muscle tolerance determined from Study S17157-00 at 18 days after administration. The application site fat UTLs were less than the previously determined safety number (1800 ppb) at 21 days after administration.

Table IV.12. Mean (± standard deviation) Concentrations (ppb) of Parent Fluralaner in Non-Application Site Tissues from Cattle Dosed Topically with Fluralaner at a Dose of up to 3.7 mg Fluralaner/kg Body Weight

WDP[1] (days)	Remote Muscle	Liver	Renal Fat	Mesenteric Fat	Kidney	Heart	Tongue
7	146.15 ± 85.46 n = 6	875.5 ± 492.13 n = 6	1292.33 ± 753.49 n = 6	1351.17 ± 787.2 n = 6	439.5 ± 241.75 n = 6	309.17 ± 184.28 n = 6	329 ± 251.99 n = 6
14	77.73 ± 34.95 n = 6	493 ± 286.53 n = 6	601.83 ± 233.84 n = 6	664 ± 256.45 n = 6	265.5 ± 146.13 n = 6	179.73 ± 106.41 n = 6	177.83 ± 96.91 n = 6
21	32.58 ± 14.17 n = 6	215.07 ± 112.78 n = 6	335 ± 156.18 n = 6	324.83 ± 158.03 n = 6	112.38 ± 57.68 n = 6	75.02 ± 34.58 n = 6	84.4 ± 36.77 n = 6
35	20 n = 1	93.6 ± 55.72 n = 2	102.7 ± 70.95 n = 3	99.97 ± 72.83 n = 3	50.5 ± 28 n = 2	17.11 ± 14.21 n = 6	19.58 ± 16.41 n = 6
42	22.48 ± 19.41 n = 2	143.75 ± 110.66 n = 2	227.7 ± 198.41 n = 2	216.1 ± 182.29 n = 2	78.55 ± 64.28 n = 2	46.65 ± 38.4 n = 2	60.6 ± 58.55 n = 2
61	<LOQ[2]	<LOQ	47.4 n = 1	48.7 n = 1	<LOQ	10.5 n = 1	13 n = 1
79	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
112	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ

[1]WDP, Withdrawal period
[2]<LOQ, less than the limit of quantification (Muscle LOQ = 8.49 ppb; Liver LOQ = 43.1 ppb; Fat LOQ = 40.3 ppb; Kidney LOQ = 24.0 ppb)

Table IV.13. Mean (± standard deviation) Concentrations (ppb) of Parent Fluralaner in Application Site Tissues from Cattle Dosed Topically with Fluralaner at a Dose of up to 3.7 mg Fluralaner/kg Body Weight

WDP[1] (days)	APS[2] Muscle Anterior	APS Muscle Middle	APS Muscle Posterior	APS Fat Anterior	APS Fat Middle	APS Fat Posterior
7	146.93 ± 109.16 n = 6	123.8 ± 83.44 n = 6	141.9 ± 80.5 n = 6	1038.33 ± 451.26 n = 6	1048.5 ± 582.04 n = 6	1012.83 ± 564.53 n = 6
14	76.5 ± 33.43 n = 6	63.68 ± 29.22 n = 6	69.98 ± 33.57 n = 6	496.83 ± 199.83 n = 6	481.33 ± 124.44 n = 6	509 ± 139.86 n = 6
21	34.97 ± 15.08 n = 6	29.18 ± 12.43 n = 6	31.73 ± 14.12 n = 6	274 ± 129.2 n = 6	278.33 ± 107.68 n = 6	256 ± 111.38 n = 6
35	21.6 n = 1	19.8 n = 1	23.2 n = 1	80.93 ± 54.77 n = 3	98.75 ± 68.24 n = 2	77.7 ± 52.32 n = 3
42	22.41 ± 19.08 n = 2	31.2 n = 1	39.2 n = 1	197 ± 176.78 n = 2	170.2 ± 149.62 n = 2	178.7 ± 154.57 n = 2
61	<LOQ[3]	<LOQ	<LOQ	<LOQ	<LOQ	41.4 n = 1
79	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
112	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ

[1]WDP, Withdrawal period
[2]APS, Application site
[3]<LOQ, less than the limit of quantification (Muscle LOQ = 8.49 ppb; Fat LOQ = 40.3 ppb)

Conclusions: The data support a presumptive 44-day withdrawal period for cattle treated with fluralaner topical solution at a dose of up to 3.7 mg fluralaner/kg body weight applied topically in a narrow strip along the dorsal midline from the withers to the base of the tail if cattle are continuously exposed to temperatures at or above 60 °F after product administration.

2. Target Tissue and Marker Residue

   The data from Study S17157-00 indicate that, for edible cattle meat tissues, the target tissue is cattle liver and the marker residue is parent fluralaner.
   Study S17157-00 also indicates that parent fluralaner is an appropriate marker residue in cattle muscle.

3. Tolerances

   The data from Study S17157-00 support assigning a 500-ppb tolerance in cattle liver and a 350-ppb tolerance in cattle muscle.

4. Withdrawal Period

   The data from the warm-weather study (S20167-00) and cold-weather study (S19115-00) support the following temperature-based withdrawal period assignment:

   Environmental temperature affects the withdrawal period.

Cattle must not be slaughtered for human consumption within **98 days** of treatment.

If cattle are continuously exposed to temperatures at or above 60 °F after product administration, then cattle may be slaughtered for human consumption 44 days after treatment. Violative residues may result if cattle are exposed to temperatures below 60 °F after administration and are slaughtered at 44 days.

D. Analytical Method for Residues

1. Description of Analytical Method

   a. Determinative Procedure

      One gram of homogenized cattle liver or muscle is spiked with deuterated fluralaner internal standard and extracted once with 3 mL of extraction solvent (acetonitrile/water, 80/20, v/v). Extraction supernatant (600 µL) is combined with 600 µL of water, followed by sample purification using solid-phase extraction. The resulting solution is quantitatively analyzed by gradient reversed phase LC-MS/MS using positive ion multiple reaction monitoring (MRM). The following ion transitions are monitored for quantitation:

      Fluralaner: *m/z* 556 → *m/z* 400
      Fluralaner-d4: *m/z* 560 → *m/z* 400

   b. Confirmatory Procedure

      Sample extraction for the confirmatory procedure is identical to the one for the determinative procedure. The resulting solution is quantitatively analyzed by gradient reversed phase LC-MS/MS using positive ion MRM. The following ion transitions are monitored for confirmation:

      m/z 556 → *m/z* 457
      m/z 556 → *m/z* 400
      m/z 556 → *m/z* 160

2. Availability of the Method

   The validated analytical method for analysis of residues of fluralaner topical solution is on file at the Center for Veterinary Medicine, CPK1, 5001 Campus Drive, College Park, MD 20740. To obtain a copy of the analytical method, please submit a Freedom of Information request to: https://www.accessdata.fda.gov/scripts/foi/FOIRequest/requestinfo.cfm.

V. USER SAFETY

The product labeling contains the following information regarding safety to humans handling, administering, or exposed to Exzolt™ Cattle-CA1:

USER SAFETY WARNINGS: Not for use in humans. Keep out of reach of children.

This drug product is a skin and eye irritant; special care should be taken to avoid contact. Personal protective equipment should be worn, such as gloves, long sleeve shirt and pants, as well as glasses or goggles to prevent skin, eye and mucous membrane contact and/or drug absorption, while handling the product. In case of skin contact, wash with soap and water. If contact with eyes occurs, immediately rinse thoroughly with water. In case of accidental spill, immediately remove affected clothing and wash contacted skin with soap and water. In case of accidental ingestion, immediately rinse the mouth with plenty of water and seek medical advice.

Do not eat, drink, or smoke while handling the product. Wash hands thoroughly with soap and water immediately after use of the product.

The product is highly flammable. Keep away from heat, sparks, open flame or other sources of ignition.

VI. AGENCY CONCLUSIONS

The data submitted in support of this application satisfy the requirements of section 571(b) of the FD&C Act. The data demonstrate that Exzolt™ Cattle-CA1, when used according to the label, is safe and has a reasonable expectation of effectiveness for the conditions of use in the General Information Section above. Additionally, data demonstrate that residues in food products derived from species treated with Exzolt™ Cattle-CA1 will not represent a public health concern when the product is used according to the label.

A. Conditional Approval Eligibility

In 2018, the legislation reauthorizing FDA's animal drug user fee program (Animal Drug User Fee Program, or ADUFA IV) expanded the conditional approval pathway to allow certain additional new animal drugs that are not Minor Use/Minor Species (MUMS) drugs to be eligible for conditional approval. As provided in section 571(a)(1)(A)(ii) of the FD&C Act, as amended by ADUFA IV, to qualify for conditional approval, the non-MUMS new animal drug must meet the following two criteria:

1. The new animal drug is intended to treat a serious or life-threatening disease or condition OR addresses an unmet animal or human health need; AND

2. A demonstration of effectiveness would require a complex or particularly difficult study or studies.

Exzolt™ Cattle-CA1 was determined to be eligible for conditional approval for an NWS indication under these provisions because it prevents and treats a serious or life-threatening disease or condition and the demonstration of effectiveness requires a complex or particularly difficult study or studies. The tissue damage caused by *Cochliomyia hominivorax* in cattle can be serious and is often deadly to the animal. Therefore, the conditionally approved use of Exzolt™ Cattle-CA1 addresses a serious or life-threatening disease or condition. Demonstrating effectiveness would require a complex or particularly difficult study, or studies, because *C. hominivorax* has been eradicated in the United States, making it impossible to conduct studies in the United States using naturally infested animals to provide substantial evidence of

effectiveness. Additionally, there are significant animal welfare concerns when considering whether to conduct studies with this parasite.

Exzolt™ Cattle-CA1 was determined to be eligible for conditional approval for a cattle fever tick indication under these provisions because it addresses an unmet animal need and the demonstration of effectiveness requires a complex or particularly difficult study or studies. Currently there is no approved animal drug in the United States for treatment/control of *R. microplus*, which is a reportable tick in southern Texas, therefore Exzolt™ Cattle-CA1 addresses an unmet animal need. Demonstrating effectiveness would require a complex or particularly difficult study, or studies, because *R. microplus* has been eradicated in the United States with the exception of an established quarantine zone in some Texas counties that border Mexico, making it logistically challenging to conduct studies in the United States using naturally infested animals to provide substantial evidence of effectiveness.

Therefore, the FDA determined that Exzolt™ Cattle-CA1 met the eligibility criteria for conditional approval for both NWS and cattle fever tick indications.

B. Marketing Status

Exzolt™ Cattle-CA1 is conditionally approved for one year from the date of approval and is annually renewable for up to four additional one-year terms.

This product may be dispensed only by or on the order of a licensed veterinarian (Rx marketing status). Adequate directions for lay use cannot be written because professional expertise is required to monitor the safe and effective use of this product.

C. Exclusive Marketing Rights

Exzolt™ Cattle-CA1, as approved in our approval letter, does not qualify for exclusive marketing rights under section 573(c) of the FD&C Act because it is not a designated new animal drug under section 573(a) of the FD&C Act.

D. Patent Information

For current information on patents, see the Green Book Reports in the Animal Drugs @ FDA database.