v. 3, n. 10, 2023

International Journal of Biological and Natural Sciences

# NOVEL COMBINATION OF FIPRONIL (1%) AND EPRINOMECTIN (0.5%) FOR INTERNAL AND EXTERNAL PARASITE CONTROL AND ITS IMPACT ON DAILY AVERAGE WEIGHT GAIN ON BEEF CATTLE, FED IN JALISCO, MEXICO

*Jose Alberto Erales Villamil*
Universidad Autonoma de Yucatan, Facultad de Medicina Veterinaria y Zootecnia, Merida Yucatán, Mexico
Orcid: 0001-7326-8903

*Josue Gomez Estrada*
Private Practice, Guadalajara Jalisco, Mexico

*Marcela Viveros*
Private Practice, Guadalajara Jalisco, Mexico

**Abstract:** Parasite control on beef cattle is necessary to ensure health and good performance. Our chemical tools to control parasites sometimes put at risk the environment, animal workers or endanger natural resources such as fresh water or soil; other times are a source of stress and pain for the animals when they are applied by immersion, sprayed, or injected, respectively. A novel combination of Fipronil 1% plus Eprinomectin 0.5% applied pour-on could prevent environmental over exposure and reduce stress or pain in the animals. 299 male and female growing beef cattle were followed over 116-day period and the use of this novel combination was compared to the conventional Moxidectin 1% injected treatment. ADG was increased in 0.063kg / head in 149 animals that received Fipronil 1% plus Eprinomectin 0.5%. Gender or Breed interactions with treatment did not influence ADG. Novel combination of Fipronil 1% plus Eprinomectin 0.5% provided internal and external parasite control and increased ADG when compared to Moxidectin 1% use, under feedlot conditions in Jalisco, Mexico.
**Keywords:** Feedlot, beef cattle, ADG, parasites, Mexico.

## INTRODUCTION

Blood and fluids loss due to external and internal parasites are responsible for major economic losses (Rodriguez et al., 2017) Internal parasites also damage mucosal tissue increasing fluid, electrolyte and nutrient loss into the lumen and triggering an inflammatory response which results in an animal with decreased appetite and decreased gut retention time (Stromberg and Gasbarre, 2006). Major economic losses in global beef production are due to clinical and subclinical health problems; some authors estimate this loss to be around $43.57 USD per animal per year (Rodríguez et al., 2017; Esteve-Gassent et al., 2016; Grisi et al., 2014). Environmental conditions determine epidemiology of internal and external parasites (Charlier et al., 2020), in México, geographic and climatic conditions, in important cattle production regions, favor parasites infestations making parasitosis one of the most important health problem in cattle production. Helminths, flies, ticks, and blood-parasites they transmit, compose the greatest risk for health in every cattle operation to preserve health and expected production efficiency.

Parasite control, in domestic animals, is regularly achieved by a combination of strategies such as the use of antiparasitic chemicals. These chemicals can be sprayed, injected, poured-on, or the animal can be immersed into it. (Miller et al., 2013; Rodríguez et al., 2014; Jonsson and Hope, 2007). Some chemicals require to be diluted to be applied to the animal. Such products are used sprayed or for immersion baths. Cattle dips uses huge amounts of water (around 12,000 liters) to dilute the chemical and once the cattle dip receives organic material such as dirt and manure, reduces its effectiveness and renewing its content is necessary to remain effective. Disposal of cattle dips, as well as water used to dilute sprayed chemicals become a concern for contamination of underground water and reduction in availability of fresh clean water.

For many decades, parasite control has been applied to reduce the negative impact on production, but also to reduce animal stress. Parasites and treatments are important sources of stress in cattle production, external parasites' constant bites provoke uneasiness and pain, besides blood loss. Regular treatments by injection, immersion and spraying are also sources of stress due to the management and process of cattle. Deworming and external parasite control increase animal welfare (Love, et al., 2017) and this can be enhanced if the application of the antiparasitic is poured-on and requires no excessive management. The use of pour-on products reduces the risk of negative impact on the environment, preserves water resources and reduces the stress of dipping, spraying, or injecting the animal.

Integrated parasite control strategies that preserve water resources and reduce the risk of environment contamination and animal workers exposure are needed. A pour on commercial product combining Fipronil (1%) and Eprinomectin (0.5%) is expected to have reduced water resource usage, reduced environmental contamination, reduced animal workers exposure, internal and external parasite loads controlled, reduce the negative effects of management and external parasites stress on the animal and thus an important positive impact on average daily weight gain (ADG) on cattle fed in confinement, under feedlot conditions in the tropics of Mexico.

## METHODS

A field trial was set to test the impact of Fipronil 1% plus Eprinomectin 0.5% pour-on used in feedlot animals fed in Jalisco, Mexico to control parasite loads and increase ADG. Regularly, feedlots in Mexico receive male and female animals weighting anywhere from 180kg to 400kgs and process them to final market weight around 550 to 600 kg. Animals are classified into one of three different racial groups, (European, Zebu and Cross breeds of European and Zebu). Diets offered are high protein - high energy integral diets to promote the highest ADG possible.

## SAMPLE SIZE

To identify differences as small as 0.060 kg/day/head at a 95% confidence (1-α) and an 80% power (1-β), a sample size was obtained using the software MedCalc Ver. 10.1 of 142 animals per group with an estimated standard deviation of 0.180 kg/day/head in both groups ($p \leq 0.05$).

## PROCEDURES

328 animals received in the feedlot, from February 27th to 29th, 2020, were included in the trial and processed according to the regular Standard Operating Procedures at the feedlot, except for the antiparasitic treatment. 169 male and female beef cattle were received on February 27th and treated with a novel combination of fipronil (1%) and eprinomectin (0.5%) pour -on. On February 28th and 29th, 159 male and female beef cattle were received and treated conventionally with an injectable antiparasitic (moxidectin 1%). Antiparasitic treatments were dosed as labeled. Animals were allocated in groups (pens) according to their weight and gender and irrespective to their anti-parasite treatment. Mortalities and animals moved to a sick pen were no longer followed up in this trial. All other treatments and husbandry were identical for the animals and animal workers were not aware of any difference in treatments.

Animals were weighted individually every two months, and weights recorded on a computer file. No fasting period was observed. ADG was computed by subtracting the initial weight to the actual weight and dividing the result by the number of days between weights. Measures and ADG was kept on the computer file until follow up period ended.

## STATISTICAL ANALYSIS

Descriptive statistic for group estimates, and a "*One-tail T test*" to compare ADG means were performed. Two-way Anova was used to identify if any other variable could bias the results, as described by Dohoo et al., 2009 and 2012. StatCrunch software by Pearson™ was used to analyze the data.

## ETHICAL STATEMENT

No harm was inflected on these animals as a result of the trial, its treatments, or procedures.

## RESULTS

328 animals were included and followed over a mean period of 116 days (86 to 158 days, depending on their initial weight) until market weight. Censored data were removed resulting in "Conventional" treatment group size of 150 animals and Novel combination of Fipronil + Eprinomectin group size of 149 animals.

## GENDER

| **Frequency table results for Sex:** Group: Tx=Conventional Count = 150 | | | Group: Tx=Fipronil + Eprinomectin Count = 149 | | |
|---|---|---|---|---|---|
| **Sex** | **Frequency** | **% of Total** | **Sex** | **Frequency** | **% of Total** |
| **F** | 51 | 34.0 | **F** | 48 | 32.2 |
| **M** | 99 | 66.0 | **M** | 101 | 67.8 |

Table #1 Gender composition for treatment groups.

Each treatment group included male and female individuals in similar proportion. Although gender had a significant effect on daily ADG and mean difference (0.247kg/day/head) in male vs female were statistically significant ($p<0.001$); there were no statistically significant differences for the interaction of Male or Female by treatment, thus gender did not influence the ADG per treatment. ($p=0.072$). Table #1 shows the gender proportion (sex) for each group and table #2 presents the means and counts per gender and treatment. In figure #1, we can observe the differences in means for the interaction of gender and treatment.

| | **F** | **M** | |
|---|---|---|---|
| **C** | 1.460 (51) | 1.723 (99) | 1.634 (150) |
| **F+E** | 1.542 (48) | 1.770 (101) | 1.697 (149) |
| | 1.500 (99) | 1.747 (200) | 1.665 (299) |

Table #2. ADG and animal counts, in parenthesis, per gender and treatment.

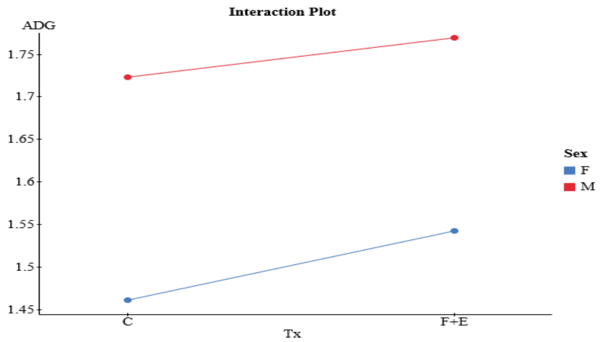


Figure #1. ADG per treatment group for male and female animals.

Although male and females ADG are significantly different *($p< 0.001$)*, no differences per treatment when compared males *($p = 0.665$)* or females *($p=0.497$)* were observed.

## BREEDS

Both groups had a combination of Zebu, European and Crossbred animals included in different proportions. Breed classification was performed by feedlot workers according to external breed characteristics of each animal. These proportions of breed group inclusion are presented in table #3.

| Frequency table results for Breed: **Group: Tx=Conventional** Count = 150 | | | **Group: Tx=Fipronil + Eprinomectin** Count =149 | | |
|---|---|---|---|---|---|
| **Breed** | **Fre-quency** | **% of Total** | **Breed** | **Fre-quency** | **% of Total** |
| **ZEBU** | 38 | 25.3 | **ZEBU** | 22 | 14.8 |
| **CZ EUR** | 101 | 67.3 | **CZ EUR** | 26 | 17.4 |
| **EUROPE** | 11 | 7.3 | **EUROPE** | 101 | 67.8 |

Table #3 Breed group inclusion per treatment.

Breed effects were analyzed using a two-way Anova test to see if the interaction of breed group influenced the overall ADG, finding no statistical significance *(p=0.488)* for the interaction between Treatment and Breed. In table #4 ADG per breed and treatment are presented.

| | CZ EUR | EUROPE | ZEB | |
|---|---|---|---|---|
| C | 1.627 (101) | 1.676 (11) | 1.641 (38) | 1.634 (150) |
| F+E | 1.623 (26) | 1.701 (101) | 1.765 (22) | 1.697 (149) |
| | 1.626 (127) | 1.698 (112) | 1.686 (60) | 1.665 (299) |

Table #4. ADG means and counts per Breed and Treatment.

Outcome of interest was ADG, means estimated were 1.634 kg/day/head for the conventional treatment and 1.697 kg/day/head, for the novel combination of Fipronil and Eprinomectin, obtaining 63g more per head every day, than the Moxidectin group (Tables #2 and #4). This difference was statistically significant at 95% confidence level (p=0.041; One-tail "T" test).

## DISCUSSION

This study included male and female beef cattle which have different ADG. Ideally, male and female should be studied and analyzed separately, however in doing so, our sample size available would be reduced to a point where no results would be robust enough to be significant at the specified confidence level. Male and Female analysis was possible because proportions were similar in both treatment groups (see table #2) and hence, did not bias the results.

Breed groups were also a concern in analyzing the data, since each treatment group had different proportions of breeds included. Zebu cattle (*Bos indicus)* are known to have smaller ADG when compared to European breeds (*Bos taurus*) however, European breeds are more susceptible to parasites and warm climates reduces their feed intake and thus, their ADG. No environmental indicators were measured (temperature or humidity) nor parasite loads were monitored, however a two-way Anova was run to analyze if breed group and treatment interactions could have biased the results, and no evidence was found for that (See table #4).

Differences in ADG observed in this study might encourage the use of this innovations which provides an effective parasite control (Gomez and Erales, 2020), promotes the preservation of water resources and reduces stress due to injections, spraying or dipping the animals, contributing to an overall animal welfare.

Present study analyzes the use of this novel combination of Fipronil and Eprinomectin under feedlot conditions in Mexico but should also be tested on grazing animals and over longer periods of time, to fully measure the net benefits over animal production and welfare and on water savings, decreased contamination, and decreased exposure to chemicals for animal workers.

## CONCLUSIONS

The use of a combination of Fipronil (1%) and Eprinomectin (0.5%) applied pour-on to control parasites, positively impacted ADG on beef cattle under feedlot conditions in Mexico. A 0.063kg increase in ADG resulted in 7.3kg more, over a 116-day period, per head, when Fipronil 1% + Eprinomectin 0.5% pour- on was used, compared to injectable moxidectin (1%).

## FUNDING

This research was funded by Boehringer Ingelheim Animal Health, Mexico.

## CONFLICT OF INTEREST

The authors declare no conflict of interest.

## REFERENCES


1. Charlier, J., Höglund, J., Morgan E.R., Geldhof P., Vercruysse J., Claerebout, E. (2020) **Biology and Epidemiology of gastrointestinal Nematodes in cattle**. Vet Clin Food Anim 36 (2020) 1-15.

2. Dohoo I., Martin W., Stryhn H. (2009) **Veterinary Epidemiologic Research** 2nd Ed. ISBN 978-0-919013-60-5 VER Inc. WWW.upei.ca/ver

3. Dohoo I., Martin W., Stryhn H. (2012) **Methods in Epidemiologic Research**. ISBN 978-0-919013-73-5 VER Inc. WWW.upei.ca/ver

4. Esteve-Gassent M.A., Castro Arellano I., Feria Arroyo T.P., Patino R., Li A.Y., Medina R.F., Perez de Leon A.A., Rodriguez Vivas R.I. (2016) **Translating Ecology, Physiology Biochemistry, and population genetics research to meet the challenge of tick and tick-borne diseases in North America**. Archives of Insect Biochemistry and Physiology, Vol 92, No.1, 38-64

5. Gomez E. J., Erales V. J., (2020) **"Prueba de efectividad de Duoline (Fipronil 1% + Eprinomectina 0.5% contra mosca del cuerno y parásitos gastroentéricos en Mexico**". *Datos no publicados.*

6. Grisi L., Leite R.C., de Souza J.R., Medeiros A.T., Andreotti R., Duarte P.H., Perez de Leon A.A., Barros J., Silva H. (2014) **Reassessment of the potential economic impact of cattle parasites in Brazil**. Braz. J. Parasitol. Jaboticabal v. 23, n.2, p.150 -156.

7. Jonsson N.N., Hope M. (2007) **Progress in the epidemiology and diagnosis of amitraz resistance in the cattle tick *Boophilus microplus***. Veterinary Parasitology. 146(2007)193-198.

8. Love, J.W., Kelly, L.A., Lester, H.E., Nanjiani, I., Taylor M.A., Robertson C., (2017) **Investigating anthelmintic efficacy against gastrointestinal nematodes in Cattle by considering appropriate probability distributions for fecal egg count data**. International journal for Parasitology. Drugs and Drug Resistance. 7 (2017) 71-82. (https://doi.org/10.1016/j.ijpddr.2017.01.002)

9. MedCalc (2009) Version 10.4.8.0 **Statistics Software** Copyright 1993-2009 Frank Schoonjans.

10. Miller R.J., Almazan C., Ortiz M., Davey R.B., George J.E., Perez de Leon A. (2013) **First report of fipronil resistance in *Rhipicephalus (Boophilus) microplus* of Mexico**. Veterinary Parasitology 191 (2013) 97-101.

11. Pearson (2020) **StatCrunch software**. Copyright 2019 Pearson Education Inc.

12. Rodriguez R.I., Perez L.C., Rosado J.A., Ojeda M.C., Martinez I., Miller R.J., Li A.Y., Perez de Leon A., Guerrero F., Klafke G. (2014) **Rhipicephalus (Boophilus) microplus resistant to acaricides and ivermectin in cattle farms of Mexico**. Braz. J. Vet. Parasitol., Jaboticabal, V.23 n. 2, p113 -122.

13. Rodriguez Vivas, R.I., Grisi, L., Perez de Leon, A.A., Silva Villela, H., Torres Acosta, F.J., Fragoso Sanchez, H., Romero Salas, D., Rosario Cruz, R., Saldierna, F., Garcia Carrasco, D. (2017) **Potential economic impact assessment for cattle parasites in Mexico. Review.** Rev Mex Cienc Pecu 2017; 8 (1) 61 – 74. (http://dx.doi.org/10.22319/rmcp.v8i1. 4305)

14. Stromberg, B.E.; Gasbarre, L.C. (2006) **Gastrointestinal Nematode control programs with emphasis on cattle**. Vet Clin Food Anim 22 (2006) 543 – 565.

## STAT ANNEX

Two Way Analysis of Variance results:

Responses: ADG

Row factor: Tx

Column factor: Sex

ANOVA table, Unbalanced.

| Source | DF | Type III SS | MS | F-Stat | P-value |
|---|---|---|---|---|---|
| Tx | 1 | 0.27281683 | 0.27281683 | 3.2547951 | 0.0722 |
| Sex | 1 | 3.982141 | 3.982141 | 47.50826 | <0.0001 |
| Interaction | 1 | 0.020379155 | 0.020379155 | 0.24313007 | 0.6223 |
| Model | 3 | 4.3067993 | 1.4355998 | 17.127181 | <0.0001 |
| Error | 295 | 24.726892 | 0.083819971 | | |
| Total | 298 | 29.033691 | | | |

Means and counts table.

| | F | M | |
|---|---|---|---|
| C | 1.4604118 (51) | 1.7232626 (99) | 1.6338933 (150) |
| F+E | 1.5421667 (48) | 1.7699208 (101) | 1.6965503 (149) |
| | 1.5000505 (99) | 1.746825 (200) | 1.6651171 (299) |

Tukey HSD results (95% level) for Tx:

C subtracted from

| | Difference | Lower | Upper | P-value |
|---|---|---|---|---|
| F+E | 0.062657002 | -0.0032458316 | 0.12855984 | 0.0623 |

Tukey HSD results (95% level) for Sex:

F subtracted from

| | Difference | Lower | Upper | P-value |
|---|---|---|---|---|
| M | 0.24677449 | 0.17675641 | 0.31679258 | <0.0001 |

Tukey HSD results (95% level) for Tx*Sex:

C,F subtracted from

| | Difference | Lower | Upper | P-value |
|---|---|---|---|---|
| C,M | 0.26285086 | 0.13391773 | 0.391784 | <0.0001 |
| F+E,F | 0.081754902 | -0.068674763 | 0.23218457 | 0.4977 |
| F+E,M | 0.30950903 | 0.18101066 | 0.4380074 | <0.0001 |

C,M subtracted from

| | Difference | Lower | Upper | P-value |
|---|---|---|---|---|
| F+E,F | -0.18109596 | -0.31266147 | -0.049530451 | 0.0025 |
| F+E,M | 0.046658166 | -0.05913517 | 0.1524515 | 0.6654 |

F+E,F subtracted from

| | Difference | Lower | Upper | P-value |
|---|---|---|---|---|
| F+E,M | 0.22775413 | 0.096614655 | 0.3588936 | <0.0001 |

Two Way Analysis of Variance results:
Responses: ADG
Row factor: Tx
Column factor: Breed
ANOVA table, Unbalanced.

| Source | DF | Type III SS | MS | F-Stat | P-value |
|---|---|---|---|---|---|
| Tx | 1 | 0.093935598 | 0.093935598 | 0.96683286 | 0.3263 |
| Breed | 2 | 0.23612343 | 0.11806172 | 1.2151511 | 0.2982 |
| Interaction | 2 | 0.13946682 | 0.06973341 | 0.71773166 | 0.4887 |
| Model | 5 | 0.56638158 | 0.11327632 | 1.1658974 | 0.326 |
| Error | 293 | 28.467309 | 0.097158052 | | |
| Total | 298 | 29.033691 | | | |

Tukey HSD results (95% level) for Tx:

C subtracted from

| | Difference | Lower | Upper | P-value |
|---|---|---|---|---|
| F+E | 0.062657002 | -0.008297825 | 0.13361183 | 0.0833 |

Tukey HSD results (95% level) for Breed:

CZ EUR subtracted from

| | Difference | Lower | Upper | P-value |
|---|---|---|---|---|
| EUROPE | 0.072603346 | -0.02257651 | 0.1677832 | 0.1724 |
| ZEB | 0.060545013 | -0.054482118 | 0.17557214 | 0.4306 |

EUROPE subtracted from

| | Difference | Lower | Upper | P-value |
|---|---|---|---|---|
| ZEB | -0.012058333 | -0.12953093 | 0.10541426 | 0.9683 |

Tukey HSD results (95% level) for Tx*Breed:

C,CZ EUR subtracted from

| | Difference | Lower | Upper | P-value |
|---|---|---|---|---|
| C,EUROPE | 0.049577858 | -0.23432743 | 0.33348315 | 0.9961 |
| C,ZEB | 0.014422355 | -0.15574507 | 0.18458978 | 0.9999 |
| F+E,CZ EUR | -0.0040654989 | -0.20070737 | 0.19257637 | 1 |
| F+E,EUROPE | 0.074188119 | -0.051639453 | 0.20001569 | 0.5386 |
| F+E,ZEB | 0.13794149 | -0.072437335 | 0.34832032 | 0.416 |

C,EUROPE subtracted from

| | Difference | Lower | Upper | P-value |
|---|---|---|---|---|
| C,ZEB | -0.035155502 | -0.34130349 | 0.27099248 | 0.9995 |
| F+E,CZ EUR | -0.053643357 | -0.37526062 | 0.26797391 | 0.9969 |

| | Difference | Lower | Upper | P-value |
|---|---|---|---|---|
| F+E,EUROPE | 0.024610261 | -0.25929503 | 0.30851555 | 0.9999 |
| F+E,ZEB | 0.088363636 | -0.24183159 | 0.41855886 | 0.9727 |

C,ZEB subtracted from

| | Difference | Lower | Upper | P-value |
|---|---|---|---|---|
| F+E,CZ EUR | -0.018487854 | -0.24606729 | 0.20909158 | 0.9999 |
| F+E,EUROPE | 0.059765763 | -0.11040166 | 0.22993319 | 0.9152 |
| F+E,ZEB | 0.12351914 | -0.11602966 | 0.36306793 | 0.6779 |

F+E,CZ EUR subtracted from

| | Difference | Lower | Upper | P-value |
|---|---|---|---|---|
| F+E,EUROPE | 0.078253618 | -0.11838826 | 0.27489549 | 0.8636 |
| F+E,ZEB | 0.14200699 | -0.11701945 | 0.40103344 | 0.6172 |

F+E,EUROPE subtracted from

| | Difference | Lower | Upper | P-value |
|---|---|---|---|---|
| F+E,ZEB | 0.063753375 | -0.14662545 | 0.2741322 | 0.9535 |