

PONENCIAS MAGISTRALES

## The “Art of War” against gastrointestinal nematodes in sheep and goat herds of the tropics

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### Abstract

The present work delves into the concept of infections by gastrointestinal nematodes (GIN) in ruminants in the light of new findings of the animal-parasite-vegetation relationship and shows how to use these to guide the rational use of alternative control methods. First, we reflect on the control of the GIN in the current era and how the indiscriminate use of anthelmintics (AH) has generated a big problem of resistance to these drugs. The research on AH-resistant GIN helped to recognize that high GIN burdens are found in a low proportion of animals in each herd. This makes it possible to propose a new control paradigm based on the selective use of AH only in those animals that need treatment. It is proposed that low GIN infections in herds are due to: (i) the use of native GIN-resistant breeds, (ii) the low infectivity of grasslands for many months of the year, (iii) the consumption of native tropical plants containing

secondary compounds (SC) affecting several stages of the GIN cycle, and (iv) grazing behaviour that limits the consumption of infective phases of GIN in low-rise fodder at hours of increased infectivity. There is a need to use a targeted selective treatment strategy aiming to reduce false positives and false negatives events commonly found in several strategies. To reduce reliance on conventional AH, alternative control methods affecting GIN phases outside or inside the host are required. Possibly many producers are already using some alternative method of control without being aware of this. For example, the use of tropical breeds takes advantage of their enhanced ability to resist GINs. In addition, browsing in the tropical forest vegetation involves consuming nutraceutical plants that provide nutrients and SC with AH activity. The aforementioned strategies can be reinforced with dietary supplementation to improve

productivity and immune response against GINs. Some producers might be interested in rotational grazing, which serves to evade the infecting larvae in the pastures. In the future they may have access to nematophagous fungi that can be used to prevent L3 larvae from leaving the faeces and contaminate the fodder. One element that will be important is the Barbervax<sup>®</sup> vaccine that uses an antigen obtained from the *Haemonchus contortus* intestine to generate antibodies against that parasite achieving parasitic burdens reductions > 90%. In conclusion, it is necessary to continue deepening the animal-parasite-vegetation relationship in order to be led by such knowledge to make better decisions about control methods. All this to allow the sustainability of the GIN control strategy in each herd.

**Keywords:** Post-anthelmintic era. Alternative control methods. Combined control strategies.

## Introduction

A principle of the ancient book "The Art of War", written by Sun Tsu between the 6th and 5th centuries BC, suggests "to avoid as far as possible the use of weapons to destroy the enemy" (Sun Tsu, 1999). However, in the war against gastrointestinal nematodes (GIN) of domestic ruminants we have done exactly the opposite of that old recommendation. Humanity has used anthelmintics (AH) as the main and sometimes only weapon in the fight against GIN. As would have been anticipated in that ancient Chinese book, the parasites are winning the war based solely on AH. The GIN used an ancient tool of nature: the selection of populations genetically adapted to resist the AH. These drugs have ceased to work in a large proportion of herds, mainly in tropical areas, as observed in Brazil (Salgado and Santos, 2016) and Mexico (Herrera-Manzanilla et al., 2016). Consequently, a change in the GIN's control paradigm is required. Such change was proposed almost two decades ago in Mexico (Torres-Acosta, 2000). At that time, the AH resistance situation in Mexico was unknown and very few alternative control methods had been tested. By the beginning of the next decade there was already a large amount

of studies suggesting an alarming situation of AH-resistant GIN in the Americas (Torres-Acosta et al., 2012), and these results seem to worsen over time (Sepúlveda-Vázquez et al., 2017). In the same period, it has been demonstrated that several alternative methods of GIN control are feasible under tropical conditions (Torres-Acosta et al., 2014a). Several of these control methods attack the free-living stages of GIN in pastures, and others attack GIN inside animals. The new GIN control paradigm must be based on the use of different control methods used in a simultaneous and rational manner. However, for the new paradigm become successful it is still necessary to accomplish another old Sun Tsu maxim (1999): "to know the enemy well (GIN species, their seasonality, etc.) and to know yourself" (the control strategies you're going to implement, the farms, the animals, the paddocks). In general, we think we know the GIN "very well", but we have not really deepened on our knowledge about them. As for the knowledge of ourselves, perhaps we know something about the farms or animal breeds, but we may not be able to recognize the weaknesses and strengths of each of the existing alternative GIN control methods, or their potential antagonisms or synergies. We must be completely aware of the advantages and disadvantages of each of the GIN control methods in order to use them correctly. In addition, we must understand that the different control methods must be used within a well-grounded strategy since "the tactics without defined strategy are the noise before defeat" (Sun Tsu, 1999). This paper delves into some concepts of GIN infections in ruminants in the light of new findings of the animal-parasite-vegetation relationship and shows how to use knowledge of this relationship to guide the rational use of alternative control methods.

## Thematic development

### A new paradigm for GIN control in domestic ruminants

Continuing with the teachings of Sun Tsu (1999), "weapons are fatal instruments that must be used when there is no other alternative". However, once

humanity had access to modern conventional AH, with efficiencies over 95% reduction against GIN, virtually everyone involved in ruminant production abandoned any GIN control method they would have used before that time in history. The "AH era" was born as a turning point for humanity, in which these drugs were considered as the only weapon of reliable control against GINs, able to improve productivity and prevent clinical signs of GIN in grazing animals. The use of conventional AH is still the paradigm of modern GIN control. However, AHs were used inappropriately and excessively, resulting in the fact that, at the beginning of the second decade of the twenty-first century, the sustainability of AH-based GIN control in small ruminant herds of the present and future is questioned.

The different AH resistance field surveys have generated information in two valuable aspects. The first salient result is the high frequency of herds with AH-resistant GIN in different parts of the world, which records the alarming situation of herds in which GIN are resistant to one or more classes of AH. These results make it clear that the irrational use of AH is ushering in a new era in the control of GIN: "the post-anthelmintic era". This is defined as the historical moment of each farm in which treatments with AH become unnecessary because its populations of GIN are multi-resistant to all kinds of AH, both individually or in any combination. Both in Brazil and Mexico there are already reports of herds in which none of the available AH classes manage to control the GIN (Silva et al., 2018). The second salient product of those surveys, and perhaps even more valuable, is the knowledge of the quantity of animals in each herd showing low eliminations of GIN eggs per gram of faeces (EPG). It is well known that the faecal EPG excretion gives us a very accurate idea of the amount of GIN inside animals, where a low faecal EPG excretion is associated with low GIN burden in small ruminants, and high faecal EPG excretion is associated with high GIN burden in the animal (Mohammed et al., 2016). The latter shows that in each herd surveyed there were very few animals with high GIN burdens, and these animals coexist with a large majority of animals carrying low parasite burdens (Table 1).

This phenomenon in which half of the animals have < 200 EPG has been demonstrated in goats,

sheep and cattle from different tropical areas of Mexico (Torres-Acosta et al., 2014b; Medina-Pérez et al., 2015; Soto-Barrientos et al., 2018), and is being searched in other tropical areas of Latin America. Both results support the need to change the paradigm of GIN control in domestic ruminant farms. On the one hand, there is an urgent need to reduce our dependence on conventional AH and, on the other hand, it is also evident that there are many animals that do not need to be de-wormed as they naturally have low burdens of these parasites.

### **Why only few animals show high GIN burdens?**

The previous section suggests that, for decades, veterinarians and producers used superficial information about the reality of GIN parasitic infections in ruminants under humid and sub-humid tropical areas. The GIN infections were seen as an imminent danger for all grazing animals (Box 1). Faced with this scenario of fear, conventional AHs emerged in the second half of the 20th century (70s and 80s) as a tool to be able to have more animals per hectare, avoiding the negative effects of GINs such as animal suffering, low productivity and mortality. For many producers and veterinarians, find an animal showing any sign associated with GIN infections (diarrhoea, weakness, anaemia, low body condition, sub-mandibular oedema, etc.) was sufficient evidence to deworm all animals in a flock. New information available on the dynamics of GIN infections in grazing ruminants in humid warm tropical areas has enabled us to update and refine our concepts about GIN infections and its control (Box 1). It has been shown that GIN infections are only risky in a small proportion of the herd. In fact, these low GIN burdens in most animals could be a demonstration of mutual adaptation between parasites and their hosts, as it does not benefit the former to kill their hosts. Only when other factors affect this balance (such as malnutrition or parturition) do animals with problems arise because they cannot cover the nutritional cost of their parasites. It is therefore necessary to develop control strategies aimed at limiting GIN only in that small proportion of animals in each herd.

**Table 1** - Distribution of the amount of eggs per gram of faeces (EPG) eliminated by sheep or goats from warm and temperate tropical areas, where it is evident that 50% of the sampled population (median) has only from 50 to 200 EPG, 75% of the animals (3rd quartile) eliminated between 200 and 650 EPG, and the proportion of animals shedding more than 1000 EPG was between 2.8% and 27.9%

	Minimum	Median	3rd quartile	1000 EPG or higher	Reference
Warm regions					
Tabasco (n = 900 sheep)	0	100	550	16.8%	Medina-Pérez et al., 2015
Campeche n = 907 sheep	0	100	350	11.8%	Sepúlveda-Vázquez et al., 2017
Yucatán n = 1500 goats	0	100	650	27.9%	Torres-Acosta et al., 2014
Yucatán n = 2788 sheep	0	100	650	15%	Soto-Barrientos et al., 2018
Temperate regions					
Querétaro n = 299 goats	0	200	450	9.6%	Martinez-Ortíz de Montellano*
Querétaro n = 141goats	0	100	200	1.4%	Martinez-Ortíz de Montellano*
Morelos n = 942 sheep	0	50	300	9.8%	Martinez-Ortíz de Montellano*
Morelos n = 906 sheep	0	150	400	8.8%	Martinez-Ortíz de Montellano*

Note: \* Unpublished data.

**Box 1** - Producers and veterinarians controlling gastrointestinal nematode (GIN) infections in domestic ruminant populations grazing in tropical areas must change from current misconceptions (I) towards adopting new concepts (II) that will allow a more sustainable control of these parasites

(I) Misconceptions about GIN infections in ruminants under tropical conditions	(II) New concepts about GIN infections in ruminants under tropical conditions
<p><b>(a)</b> Grazing animals are always ingesting a large number of infective larvae (L3) of different GIN species with their pasture.</p> <p><b>(b)</b> All animals with mixed GIN infections are sick.</p> <p><b>(c)</b> All animals infected with GIN have low production or die compared to non-infected animals.</p> <p><b>(d)</b> Treatment with an effective AH always benefit health and production of infected animals.</p>	<p><b>(a)</b> Grazing animals are mainly infected during rainy season with varying amounts of L<sub>3</sub> from different GIN species.</p> <p><b>(b)</b> Only animals with severe mixed GIN infections have signs of parasitic disease.</p> <p><b>(c)</b> Only the low proportion of heavily infected animals have low production or die from infection.</p> <p><b>(d)</b> An effective AH treatment does not benefit animals with low parasite burdens.</p> <p><b>(e)</b> Animals with moderate GIN burdens are equally healthy and productive as GIN-free animals.</p> <p><b>(f)</b> It is not necessary to keep grazing animals free of GIN by means of suppressive AH treatment.</p> <p><b>(g)</b> Uncleaned animals serve as a refuge of susceptibility to AH for the farm especially during the dry season.</p> <p><b>(h)</b> During the dry season the AH treatments should be only to those animals which really need it.</p>

The low number of animals with high GIN burdens on farms from warm tropical areas may be due to different factors:

(i) *Tropical animal breeds have a great ability to demonstrate an innate response or an acquired immune response against GINs.* These results in animals of different ages with low EPG excretions compared to animals of breeds less adapted to GIN, even when they are exposed to similar levels of pasture infectivity (Palomo-Couoh et al., 2016, 2017).

(ii) *Some herds in humid warm tropical areas may have low levels of GIN infective larvae in their*

*paddocks.* The GIN infectivity in the vegetation of warm tropical zones has been scarcely studied. The conditions in the humid and sub-humid warm tropics have always been considered ideal for the survival of GIN free-living life stages. However, the use of tracer animals grazing the tropical rainforest of Yucatan, Mexico, showed that this vegetation has low or no GIN infective larvae (L3) during the dry season (February to May) (Torres-Acosta et al., 2006), while the amount of L3 in the foliage gradually increases along the rainy season, being scarce during June and July, and increases in the

months of August to December (Jaimez-Rodríguez et al., 2019). Although it is very likely that the dry season infectivity is generally low in warm tropical areas, GIN infectivity during the rainy season could be high in those herds with high animal density per hectare and in those herds using parasite-susceptible breeds.

(iii) *Many tropical vegetation plants contain secondary compounds (SC) that reduce GIN infection by affecting different life stages of parasites.* Many plants in tropical forests contain SC that affect L3 establishment or affect the fertility of GIN adult females. They can also limit the survival capacity of GIN eggs deposited in the faeces. Therefore, animals fed on foliage from some native plants of the tropical forest may have a lower GIN infection, reducing its EPG excretion (Méndez-Ortiz et al., 2019).

(iv) *Animals browsing the rainforests choose to consume foliage from browsing plants in the early hours of the day, while they increase their grass consumption at noon.* Animals apparently avoid consuming grass when the morning dew is present, and increase their grass consumption after dew evaporates during the hottest part of the day (Torres-Fajardo et al., 2019). This behaviour could be aimed at avoiding dewy grass, which may have more L3. It could also suggest that animals first consume plants with SC aiming to avoid the establishment of L3 and then they consume grass, which could be the strata most contaminated with L3.

### **Treat with anthelmintics only those animals that really need treatment**

In this scenario, where a large proportion of the farm's ruminant population has low GIN burdens, it is easy to imagine that we only need to deworm animals with high GIN burdens that affect their production or health, and we do not need to de-worm the other animals. That is, we must develop a methodology to help us identify when it is necessary to fight GIN and when not. Recent studies showed that the targeted selective treatment (TST) of sheep and goats is possible and allows us to maintain a high percentage of animals in the herd (> 60% of adult females) without a single AH treatment every year (Table 2).

Several TST protocols are now being studied to make this work simpler and more effective for tropical and temperate regions. Ideally, any TST methodology should be simple to perform and focus on finding animals with high EPG values. However, in some countries, TST is already applied in animals with pale mucous membranes using the FAMACHA® methodology, or with low body condition, or with weight gain below a certain threshold (Rizzon et al., 2019). These TST methods seek to avoid the need to take faecal samples for egg counting in the laboratory. However, so far these methodologies may result in several false positives results that are dewormed without needing it, and false negatives remaining without deworming (Soto-Barrientos et al., 2018).

**Table 2** - Proportion of adult ewes kept without deworming or with 1, 2, 3 and 4 or more treatments per year in different production systems maintained with targeted selective treatment combining low body condition or pale FAMACHA® and the excretion equal or higher than 750 eggs per gram of faeces (EPG) (in Tabasco EPG excretions were equal or greater than 1000 EPG were used)

Treatments/ewe/year	Commercial Yucatán <sup>1</sup> %	Backyard Yucatán <sup>2</sup> %	Commercial Tabasco <sup>3</sup> %	Commercial Tamaulipas <sup>4</sup> %
0	63.5	68.1	65.5	33.3
1	23.2	25.2	17.8	63.3
2	10.5	6.7	8.0	3.3
3	2.9	0	4.5	0
4 or more	0	0	3.9	0

Note: <sup>1</sup> Soto-Barrientos et al., 2018; <sup>2</sup> González-Ruiz et al. (unpublished results); <sup>3</sup> Medina-Pérez et al., 2015; <sup>4</sup> Zapata-Campos et al. (unpublished results).

### Using different alternative methods to strengthen the GIN control strategy

Several alternative GIN control strategies are currently available and have been tested in ovine and caprine production systems in Mexico (Torres-Acosta et al., 2014a). The different alternative methods can be divided into two groups: those that affect the stages of these parasites outside the animal (free living stages like eggs, L1, L2 and L3), and those methods affecting parasite stages within the infected animal. All these methods must be used in a well-designed strategy since "tactics without strategy are like noise before defeat" (Sun Tsu, 1999). Each producer must be able to identify the most suitable methods to use, according to the availability and needs of their farm.

In tropical areas many producers are already inadvertently using various alternative control methods against GINs. It could even be said that they are already largely achieving "to break the enemy's resistance without fighting" Sun Tsu, 1999). For example, many producers use GIN-resistant breeds (such as Pelibuey or Blackbelly), or are grazing vegetation containing SC with AH activity, or use browsing plants which, due to their height, do not contain L3 larvae. These producers can enhance their GIN control in their resistant animals by dietary supplementation, which provides energy and protein to improve the quality of the total ration. Supplementation can be used to cover the metabolic costs caused by parasites to their hosts, which are lower than traditionally thought. Supplemented animals can improve their resilience and even resistance against GIN, enhancing their production while limiting the negative signs of GIN infections (Méndez-Ortíz et al., 2019). This method works very well on farms where the nutritional level is poor or lacking in quality or quantity (Hoste et al., 2016). However, the producer must be able to identify which nutrient is limiting and which are not or even which are in excess. This is to avoid making a bad supplementation strategy that fail to benefit the respective animals (Hoste et al., 2016).

On the other hand, the consumption of nutraceutical plants provides macronutrients to support the nutrition of parasitic animals and

contain useful SC that help controlling different GIN stages as mentioned above. Therefore, animals supplemented with plants that provide a good quality diet and also contain SC with AH activity, can lead to the possibility of achieving good weight gain and good body condition, meaning, they help "to beat the enemy without combating" (Sun Tsu, 1999). In this same philosophy, producers can implement a paddock rotation scheme that limits the use of the paddock to 2 - 3 days, and allows a 30-day rest for the paddock. With this management many L3 larvae die before the animals return to the same paddock 30 days later. In addition, producers will be able to use in the future a product containing nematophagus fungi such as *Duddingtonia flagrans* as was recently released in Australia, Europe and the United States. The *D. flagrans* fungi trap > 80% of L3 inside the faeces of infected animals using their three-dimensional trapping structures to prevent L3 from leaving the faeces to infect paddocks. Both the paddock rotation and the use of nematophagus fungi are methods allowing to reach another principle suggested by Sun Tsu (1999): "Take your enemy to a point from which they cannot get out and will die before they can escape".

Another method of GIN control that is available to producers in Australia and South Africa is the Barbervax<sup>®</sup> or Wirevax<sup>®</sup> vaccine against *Haemonchus contortus*. This vaccine may serve to illustrate another principle of Sun Tsu (1999): "Use the enemy to defeat the enemy". The vaccine is made with an antigen obtained from the intestine of these parasites. Vaccinated animals produce antibodies against this protein after 3 consecutive vaccine applications (one every 3 weeks). When the parasites in the vaccinated animals feed on blood, the latter contain antibodies that affect the intestine of parasites, causing a reduction of up to 90% in the elimination of EPG in the vaccinated animals, compared to those not vaccinated. This method was already evaluated in Mexico in growing Pelibuey lambs with excellent results (Cáceres-Mejía et al, 2016). This ingenious method also illustrates another advice from Sun Tsu (1999): "Attack your enemy where he is unprepared, appear where he does not expect you".

## Conclusion

The "post-anthelmintic era" is already a clear reality in some small ruminant farms in tropical areas. To prevent more farms from entering that stage, it is necessary to adopt a new GIN control paradigm that limits our dependence on commercial AH, using them selectively. For this it is evident that there is still much to learn from the animal-parasite-vegetation interaction. Only deep knowledge of this interaction will allow us to handle and adapt GIN control methods correctly to each conditions' change to achieve sustainable control. The use of numerous methods for GIN control is complex and requires knowledge and planning. Therefore, the actual embracing of these methods by producers depends on their capacity to perceive the benefits of controlling GIN, identifying the inherent rewards for the overall condition of the herd. As veterinarians we must be able to provide strategies that use different methods in a reasonable, simple and clear way to achieve producer satisfaction.

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