**To treat or not to treat: diagnostic thresholds in subclinical helminth infections of cattle**

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**Abstract**

Helminth infections of cattle place significant burdens on livestock production and farm economic efficiency. Heavy infections are relatively easy to detect and treat with anthelmintics. However, subclinical infections have major but often hidden impacts on animals, necessitating more refined diagnostics to detect them and ideally inform farmers about the likely impact of anthelmintic treatment on animal and herd performance. Here, we review recent advances in diagnosing three major cattle helminth infections – gastrointestinal nematodes, liver flukes, and lungworms – and the search for subclinical infection thresholds to guide treatment decisions. Combining refined diagnostic thresholds with farm-specific information on grazing systems and animal history enables farmers to tailor helminth treatments to specific epidemiological circumstances, thereby limiting anthelmintic resistance and boosting agricultural efficiency and food security.

# Impact of helminth infections on food security

Parasitic infections of cattle place significant economic burdens on production systems worldwide. They negatively impact production (e.g., weight gain and milk production), resource use, nitrogen utilisation, and greenhouse gas emissions by reducing production efficiency and incurring additional costs from anthelmintic treatments, other parasite control measures, and animal mortality [1,2]. In the European Union (EU), for instance, cattle helminth infections cost an estimated € 2.1 billion annually [3]. These losses stress global food production systems that are already caught between increasingly urgent pressures. The current war in Ukraine has intensified rising food prices driven by the increasing human population, changing dietary patterns, and climate change [4,5]. Therefore, while food systems must reverse current negative impacts on biodiversity, environmental pollution, and greenhouse gas emissions [4,6], they should simultaneously aim to increase production capacity and efficiency while securing animal welfare.

Our ability to rapidly diagnose and effectively treat cattle helminth infections is vital for reducing their impacts on animal health and production efficiency. Anthelmintic drugs are a key tool for veterinarians and farmers to manage helminth infections, but their efficacy is threatened by the rising prevalence of **anthelmintic resistance** (AR; see **Glossary**). Anthelmintics have traditionally been used to treat groups of cattle, often prophylactically, in the absence of any diagnostic information. Current advice is that anthelmintic use should evolve toward targeted strategies that limit drug treatment to specific timeframes or animals, maintaining productivity without frequent whole-herd treatment [7]. Targeted/selective treatments require local epidemiological knowledge and user-friendly, cost-effective diagnostics to identify herds or animals in which parasitic infections are affecting productivity. Sensitive, evidence-based diagnostic tests are therefore urgently needed to monitor cattle helminth infections, allowing farmers and veterinarians to develop control plans that minimise parasite transmission and use anthelmintics as selectively as possible while maximising production efficiency and animal welfare.

The concept of **subclinical thresholds** in helminth infections was first introduced by Vercruysse and Claerebout [8]. Now, more than 20 years later, we review whether the proposed thresholds still apply and if the expanding diagnostic armoury has identified additional, more refined key points for anthelmintic intervention. We begin by discussing the concept of subclinical thresholds to maximise cattle productivity and limit AR by promoting targeted anthelmintic use. Next, we review existing production-based diagnostic thresholds for the three major helminth species of cattle in temperate climates – namely, the gastrointestinal nematode (GIN) *Ostertagia ostertagi*, the liver fluke *Fasciola hepatica*, and the lungworm *Dictyocaulus viviparus*. Finally, we discuss the integration of such diagnostic tests with grazing, environmental, and socioeconomic information to achieve integrated farm treatment plans for improving cow welfare and herd performance.

# System-specific subclinical thresholds guide helminth treatment decisions

Helminths are ubiquitous on pastures worldwide, and anthelmintic treatment is usually common in cattle farm management. With increasing financial pressures on farmers and the need to reduce antimicrobial use and promote environmentally friendly production methods, there is a growing demand for tools that facilitate **targeted treatment** (TT) or **targeted selective treatment** (TST) anthelmintic strategies guided by evidence-based criteria. While obvious clinical cases still occur, the vast majority of helminth-induced economic losses are caused by infections without visible clinical signs [9]. Hence, diagnostic markers are needed to define which herds or individual animals are subclinically affected and would most benefit from anthelmintic treatment [10]. These markers can be based on pathophysiological, immunological, parasitological, production or sickness behaviour parameters [11].

But what metrics determine whether treatment is appropriate? Every marker may correlate differently with production/output losses. To inform treatment decisions, test results must be linked to subclinical thresholds (i.e., cut-off values) that define the level of infection and/or host response above which production losses are likely to occur. These thresholds should ideally be defined through research for each farming system and diagnostic marker, since they can vary depending on host, pathogen, and environmental factors [12–14]. Finally, the intervention logic will depend not only on the diagnostic used but also on economic, socio-psychological and system-specific factors [15,16]. After studies have created a base of evidence, this should be translated into practical guidelines that are co-created by scientists and people working in the field.

Efforts to define test-specific subclinical thresholds have been ongoing for over two decades, but they have been hampered by the low sensitivity of common diagnostic techniques [8]. Recent advances have expanded our diagnostic repertoire and identified quantitative associations between test results and production outcomes. This will let us begin to define specific, targeted subclinical thresholds for determining appropriate treatment approaches for economically significant cattle parasites.

# Evidence-based diagnostics for gastrointestinal nematodes

*Ostertagia ostertagi, Cooperia* spp., and (in (sub)tropical areas) *Haemonchus* spp. may be considered the cattle GIN species of highest economic importance, with *O. ostertagi* of highest pathogenic importance [17] and most often associated with decreased milk yield and average daily weight gain (ADWG) [17–20]. Below, we have focused on the major GINs in temperate climate areas (*O. ostertagi* and *Cooperia* spp.), while the diagnostic markers for these infections have been divided between young animals (where both species can impact growth and weight gain) and adult animals (where the development of immunity to *Cooperia* spp. makes *O. ostertagi* the dominant species impacting productivity in adult cattle).

*Calves and heifers*

**Faecal egg counts** (FECs) remain the most conventional diagnostic for GIN infections and the method of choice for monitoring infection patterns in **first season grazing** (FSG) calves during the grazing period. While they can be assessed with unexpensive lab equipment, they correlate poorly with parasite burden and subclinical production losses [21]. Nonetheless, FECs performed before development of immunity are useful because they provide an assessment of the larval challenge during the grazing season, a critical factor for production losses [22]. Moreover, they allow monitoring of FEC reduction after treatment as an indicator of treatment failure or AR and can be combined with molecular tests assessing GIN species composition (see below). The only quantitatively supported FEC threshold remains 200 eggs per gram (EPG) at two months post-turnout (before development of immunity). In a 1998 meta-analysis in Western Europe, calf groups that exceeded this average threshold had a high risk of developing clinical parasitic gastroenteritis [23]. However, confirmation by new studies would be valuable in light of climate change and new animal husbandry methods. A recent global meta-analysis found a negative relationship between FEC and weight gain, suggesting a significant effect even at very low FEC (10-50 EPG) [24]. However, this study pools data from various geographies and cattle breeds. Since all grazing calves shed worm eggs, and calves should be exposed to some level of parasites to develop immunity [25], FEC thresholds should balance the trade-off between immunity development and weight losses. However, no such thresholds have yet been demonstrated. Further, it is widely acknowledged that rigid interpretation of FECs can be misleading because they are influenced by numerous factors including parasite species, season, acquired immunity, and faecal consistency.

Serum or plasma pepsinogen levels are considered a more reliable marker for production-based treatments where abomasal dwelling nematodes such as *O. ostertagi* are implicated [8]. Initially, a group mean pepsinogen level of 3-3.6 units tyrosine (U Tyr) was proposed for diagnosing subclinical ostertagiosis associated with reduced weight gain [8,26]. However, subsequent studies showed that this threshold instead indicates clinical infection, with subclinical weight gain reduction (and significant post-treatment weight gain) observed in calf groups with pepsinogen levels ≥ 2.5 U Tyr [27,28]. However, poor inter-laboratory standardisation of this method hinders the effective comparison of different studies [29]; therefore, a universal production threshold has not been defined. Other drawbacks of the pepsinogen test include its relatively high cost, invasive sampling method, and short optimal sampling window. Moreover, being based on a biochemical assay, it is difficult to incorporate into routine diagnostic monitoring programmes which are often based on ELISA. An anti-*O. ostertagi* antibody ELISA on serum may overcome several drawbacks of the pepsinogen assay. Merlin *et al.* found negative associations between ADWG and mean *O. ostertagi* serum ELISA results from dairy calf groups at housing, leading them to propose a group mean threshold of 0.7 optical density ratio (ODR) [13,20]. This was later confirmed in an intervention study showing significant post-treatment weight gain in calf groups exceeding this threshold [28]. However, serum pepsinogen assays and antibody ELISAs both require invasive blood sampling and are mainly suitable for testing animals at housing to evaluate past levels of exposure and inform future control strategies. Therefore, production losses incurred during the past grazing season may not be prevented.

Because growth reduction is an early consequence of GIN infection, treatment of FSG calves that fall below a given ADWG has been proposed as a simple method for promptly addressing ongoing production losses and increasing overall herd weight gain while reducing anthelmintic usage [30,31]. First, a fixed weight gain threshold in dairy calves of 750 g/day has been proposed on the basis of a proof-of-concept [31] and was then successful in a field trial in maintaining target weights while limiting anthelmintic usage [30] in groups of dairy calves. Since ADWG is affected by many animal, farm and environmental variables, ADWG thresholds must ideally be herd-, group- or even animal-specific. Merlin *et al.* tested a mid-season TST strategy, treating only FSG calves showing an ADWG below its own group mean [28]. This flexible ADWG threshold varied from 338-941 g/day among groups, and 28-75% of animals per group were consequently treated. Bates *et al.* showed that both TT and TST approaches can be based on group- or animal-specific weight gain thresholds, respectively, although some reduced growth was observed in the TST approach [32]. Importantly, three critical points can be stressed regarding such weight gain-based treatment strategies: (i) they are only relevant if GIN infection is a predominant limiting factor for growth (i.e., when nutritional requirements are met), (ii) their application is highly dependent on the presence of weighing equipment on the farm (automatic weighing could be a solution, but this technology is not yet widely available) and (iii) as they are independent of the level of egg excretion [29], the larval contamination of pastures can remain high [33].

Finally, **pasture risk assessment** can be used as a tool combining different management characteristics (grazing history, pasture management procedures, anthelmintic treatment history) [34]. It can be performed by a questionnaire/audit approach. There are also mathematical models available [35] or, as explained above, FECs early in the grazing season also provide an idea of pasture contamination with infectious larvae. Pasture risk assessment can also be combined with individual parasitological or production parameters [20]. In a two-step procedure, Merlin *et al.* first grouped FSG calves into low- and high-exposure groups by using a mathematical model that models the number of *Ostertagia* L3 generations on plots [13]. A threshold of ≥ 3 successive L3 generations allowed identification of the high-exposure groups. Next, in these high-exposure groups, the authors proposed various ADWG thresholds that balanced maximising production versus maintaining ***refugia*** [13].

*Adult cows*

Since its commercialisation in 2007, a **bulk tank milk** (BTM) *O. ostertagi* ELISAhas become a popular test for evaluating GIN exposure and potential production losses in adult dairy cows. The associations between BTM ELISA results and herd productivity indices have been widely studied [11], and an interpretation chart was developed indicating increasing milk yield losses when ODR > 0.5 [36,37]. However, subsequent studies showed that this association does not reliably predict herd-specific milk yield responses after anthelmintic treatment [38,39], as these may be influenced by many factors besides past parasite exposure such as physiological state, nutritional status, age and concurrent infections. In beef cattle, the feasibility of a meat-juice ELISA applied along the slaughter line to assess the impact of carcass weight has been shown, but it has found no uptake up to this day [40].

Evaluation of grazing management practices and immunity development (based on the time of effective contact (TEC) with GIN larvae before the first calving) has also proved useful in assessing farm-level GIN exposure and likely production impacts [18,41]. Herds with a mean TEC < 8 months were more likely to see increased milk production after treatment than were herds with a TEC ≥ 8 months [18,42]. Ravinet *et al.* used a BTM ODR cut-off of ≥ 0.74 to define adult dairy cow groups at risk of milk production losses; combining this threshold with the TEC greatly increased its specificity, demonstrating that TST strategies based on multiple carefully selected parameters can minimise both production losses and necessary anthelmintic treatments [42]. When choosing cows to be selectively treated, mixed profiles combining criteria that characterize both the cow and its herd seem to be the most relevant (e.g., treating first- and second-lactation cows in low-TEC, high-ODR herds at housing) [42].

FECs are generally not considered useful in adult cows [40]. However, some studies found correlations of FEC with milk yield [43] and production responses following anthelmintic treatment, provided that sensitive FEC methods are used [18,19]. More research is needed to confirm these relationships, establish interpretation thresholds and consider the importance of the GIN species infecting the animal/herd.

*Novel diagnostics*

As subclinical thresholds are defined for existing assays, other groups are developing novel diagnostics that leverage recent developments to increase the speed, sensitivity, and ease of GIN detection. Several new prototype or commercial FEC tests are currently being developed based on internet-connected image capture and image recognition [44].

Molecular methods utilise the ITS-2 ribosomal DNA region to differentiate GIN species [45], which is particularly useful when combined with a FEC reduction test to identify resistant worm species that survive anthelmintic treatment. Examples include a robotic PCR platform for species-specific GIN identification from eggs in cattle faeces [46]; quantitative, real-time PCR [47]; amplification, fragment analysis, and minisequencing of the ITS-2 region [48]; and deep amplicon sequencing (nemabiome barcoding) [49]. Francis & Šlapeta recently integrated the FECPAKG2 with nemabiome barcoding, thereby combining GIN quantification and species identification [50]. FEC can also be assisted via absolute quantification of GIN DNA – Baltrušis *et al.*, for instance, developed a droplet digital PCR (ddPCR) for quantifying *Ostertagia* and *Cooperia* DNA in cattle faeces [51].

There is some promise in the detection of antibodies against larval surface antigens in saliva [52], while another evolution is the monitoring of changes in animal behaviour to detect infections. GIN infections substantially affect activity patterns and decrease grazing time in grazing calves [53] and dairy cows [54]. Studies using animal-mounted accelerometers have shown that cattle motion and rumination behaviours were affected by GIN infections, but more research is needed to evaluate whether behaviour patterns can be used as reliable indicators for helminth infection [55–57].

In validating these new diagnostics, it will be critical to associate their outputs with production losses, thereby providing farmers with quantitative thresholds for planning anthelmintic treatments and grazing management. As discussed below, these associations can be subtle and multifactorial, complicating their assessment and interpretation. However, once these links are established, novel diagnostics capable of differentiating individual GIN species have the potential to revolutionise diagnostic approaches in parasite control.

# Evolving diagnostics and thresholds in liver fluke control

The liver fluke *Fasciola hepatica* is another well-known disturber of cattle productivity and immune responses [58]. *F. hepatica* occurs globally, but its prevalence is often regionally clustered due to requiring suitable environmental conditions for its snail intermediate host [59].

Commercial methods for diagnosing *F. hepatica* cattle infections include FECs, a copro-antigen detection ELISA (cELISA), and an antibody detection ELISA (abELISA) that can be used on serum, individual milk, or BTM samples. The cELISA is based on the MM3 antibody that binds to two *F. hepatica* cathepsins [60]. These antigens persist only for the lifetime of the infection, so a positive result indicates current infection. cELISA results correlate with fluke burden [60–62], but the test’s reported sensitivity was lower in infections of < 10 EPG, particularly in cattle [62,63]. DNA amplification-based diagnostics have also been described, but they have not been widely adopted for on-farm animal diagnosis [64,65]. The sensitivity of these techniques can often be improved by repeated testing or, in the case of FEC, by increasing the amount of faeces analysed [66,67]. Charlier *et al.* showed that analysing a larger amount of faeces increased sensitivity but decreased specificity for detecting animals with ≥ 10 flukes [66].

The filtration/sedimentation-based FLUKEFINDER® test was recently shown to detect all cattle with infections > 10 flukes [68]. However, a relevant question is whether fluke diagnostics can become *too* sensitive, hampering the ability to distinguish animals with production-relevant infections. Increasing sensitivity allows detection of more lightly infected animals, but from a strictly production-based view, the necessity of treating these animals may be questionable. Although poorly researched for *F. hepatica*, leaving lightly infected animals undetected (and thus untreated) may contribute to parasite *refugia*, limiting development of AR without negatively impacting productivity [69]. Therefore, a burning question is: what level of fluke burden should be considered as negatively impacting productivity? A subclinical threshold of > 30 flukes per animal was historically proposed [8], but more recent studies have revealed that even 1-10 flukes can cause production losses that increase as the fluke burden grows [66,70]. However, losses depend not only on the number of flukes present but also on the extent of liver damage (fibrosis score) they cause, which is associated with cattle breed and nutritional factors [71]. Fine-tuning the production-based diagnostic thresholds will thus require multifactorial studies that incorporate host genetics and nutritional data.

In dairy cattle, monitoring *F. hepatica* infections via BTM offers advantages in practicality and cost-efficiency [72]. To our knowledge, the first study correlating *F. hepatica* BTM abELISA results with productivity measures (milk yield and intercalving interval) was conducted in 2007 [73], leading to the development of a commercial ELISA with interpretation thresholds (**Table 1**) designed to “predict the economic impact of *F. hepatica* infection” (SVANOVIR® *F.hepatica*-Ab, INDICAL Bioscience). Negative associations were later confirmed between results from different BTM ELISAs (e.g., IDEXX® Fasciolosis Verification Test and MM3-ELISA) and herd productivity/morbidity indices (e.g., milk solids content and ketone bodies, fertility parameters, and body condition score), particularly in high-yielding herds [74–80]. Use of the test manufacturer’s established threshold from a production perspective are proposed in **Table 1**. The ongoing transition toward selective use of anthelmintics raises the question of how to identify individual animals that require treatment within a BTM-positive herd [81]. The relationship between BTM ELISA results and within-herd prevalence of fasciolosis can be variable and non-linear [72,74,82]. Nonetheless, there is generally a consistent negative relationship between milk yield and anti-*F. hepatica* antibody levels in individual milk samples [74,80], making these a promising matrix for selective flukicide treatment. The animal-level use of diagnostics subverts the traditional control approach based on whole-herd flukicide treatment at strategic periods in the year. However, since the abolishment in the EU of flukicides with a zero-withdrawal time for milk, treating whole herds at once is not economically viable in dairy herds with year-round calving. Moreover, two studies have shown that flukicide treatment of individual cows during the dry period can increase productivity while lowering the whole-herd infection level [83,84]. Developing animal-level diagnostics (and proving that their application increases economic efficacy) is therefore a critical goal. Besides the application of ELISA on individual milk samples, other rapid on-farm diagnostics could address this unmet need. Such tests are currently under development and include an antibody detection lateral flow test and a fluke egg detection test using the FECPAK system. Once development is complete, further study will be needed to define subclinical thresholds from their test results.

# The struggle against lungworm: from vigilance toward prevention

An effective commercial vaccine for lungworm (*Dictyocaulus viviparus*)has been available for over sixty years, but several limitations have restricted its widespread use [6,85]. Current lungworm management therefore largely relies on either routine prophylactic treatment of FSG calves or vigilance and treatment at the onset of clinical signs [86]. However, the unpredictability of lungworm epidemiology hampers the accurate prediction of clinical outbreaks. Moreover, significant production losses can occur even in herds with subclinical infections [86,87]. With the partial shift of lungworm infections from young to adult cattle [85], BTM ELISA has been the primary focus of many recent studies on lungworm detection. The most commonly used ELISAs detect antibodies against recombinant or native *D. viviparus* major sperm protein (MSP) in serum or milk [88,89]. A characteristic of this antigen is that only antibodies against infections with adult (male) worms are detected, and not those against larval stages. This can be an advantage in vaccinated herds because the vaccine is based on irradiated larvae. The recombinant MSP ELISA developed in Hannover is the best studied ELISA and is further discussed below. Although its sensitivity for detecting patent lungworm infections in the field via the bulk tank milk is low (50-83%), especially in cases of low herd prevalence [90,91], fortnightly applications across a grazing season have demonstrated value in (i) detecting herds with lungworm-induced production losses irrespective of clinical status and (ii) predicting the later occurrence of clinical signs [86,92,93]. These studies indicated that while the BTM ELISA’s sensitivity to detect infection is optimal at a cut-off ~0.3 ODR [86,91,93], 0.41 ODR is a more suitable cut-off to detect production losses or predict future clinical outbreaks, especially if this threshold is exceeded more than once during the grazing season [92,93] (**Table 1**). The sensitivity of this approach could be further improved by using a pooled milk sample from first-lactation heifers [90]. Together with increasingly accurate modelling approaches that can predict the peak pasture infectivity for *D. viviparus* [94], these studies show that as our diagnostic capabilities continue to grow, we can realistically aim for the development of reliable *prevention*-based treatment plans that could reduce the production losses unavoidable in traditional vigilance-based strategies.

BTM tests are not an option in young stock or beef cattle. Although sensitive lungworm diagnostics are available for faecal (detection of L1), bronchoalveolar lavage (detection of L1), or serum (antibody detection) samples [91,95,96], the minimal sample size needed to detect at least one positive animal in a farm with coughing cattle has been found to be relatively large [91].

May *et al.* found a negative association between larval excretion and individual milk yield [87], but only one milk test-day record was used to evaluate this correlation. More studies are needed to assess (i) the value of pooled faecal samples in cost reduction and (ii) the relationships with milk yield over an entire lactation and other productivity measures in young stock. Further research is also required to determine whether incorporating meteorological data and risk factor analysis intolungwormmodelling and monitoring can improve diagnostic predictive value for production impacts or clinical outbreaks.

# Dealing with polyparasitism

The diagnostics and thresholds discussed above are meant to guide treatment decisions for specific parasite infections. In field conditions, however, cattle often have mixed infections with different parasites which may compound diagnosis and treatment decisions [97] and create diagnostic dilemmas. Different diagnostics may be required for different parasitic infections (e.g., FECs or serum pepsinogen for GIN vs. the Baermann method for detection of lungworm L1 in FSG calves), the timing of diagnosis may be different (e.g., faecal examination for GIN early in the grazing season vs. liver fluke in winter) or diagnostic parameters may be confounded by co-infections (e.g., animal weight gain as a parameter for GIN treatment may not work when there is a co-infection with liver flukeor lungworm) [98]. The optimal timing for treatment may differ and/or different anthelmintics may be required (e.g., GIN vs. *Fasciola*). Helminth infections can influence in multiple and mostly unravelled ways the course of other infective (parasitic, bacterial, viral) pathogens [99,100] and hence their combined impact on production. So far, limited evidence suggests that the impacts of co-infections with different helminth species are additive rather than synergistic [73,101].

The simultaneous monitoring of different helminth infections in a single faecal or milk sample is technically possible (e.g., [102]). This, together with knowledge of local epidemiology (e.g., focal distribution of *Fasciola* infections), habitat (e.g., wet pastures or water bodies required for transmission of *Fasciola*) and specific farm conditions (e.g.,history of lungworm outbreaks) may guide the choice and the timing of specific diagnostics and treatment approaches.

# Addressing diagnostic dilemmas: towards multicriteria decision-making in helminth control

Over the past two decades, considerable progress has been made in detecting the production impacts of common helminth infections in cattle. The established correlations are typically based on observational studies, often in large study populations, and sometimes supported by intervention studies showing causal links between helminth burden and the correlated production parameters. Local and farm-level conditions, however, greatly impact the prevalence and level of parasitic co-infections and can also modify their effects on productivity. This necessitates a thorough understanding of a farm’s production system and contextual factors before the benefits of treating **subclinical infections** can be estimated. The management of beef and dairy herds is very different. Knowledge of grazing management parameters such as pasture rotation, mowing, supplemental feeding and length of the grazing period is essential for developing TT strategies [103] and identifying the best diagnostic indicators or thresholds to use. Merlin *et al.*, for instance, found that depending on grazing management, either diarrhoea score or *Ostertagia* serum ODR was the best diagnostic parameter to explain weight gain differences in FSG heifers [20]. Nutrition and host genetics are also important factors. Improved protein nutrition is known to reduce GIN parasitism and its impact in sheep [104]. In cattle, the negative association between *Ostertagia* BTM ODR and technical production efficiency was mitigated by increasing use of concentrates and roughage [105]. In another study, *O. ostertagi* BTM ODR was negatively associated with milk yield in herds with high-performance breeds, but not in herds with dual-purpose breeds [75]. The **tolerance** of beef cows to liver fluke infection differed between breeds (genetic factors) and producers, suggesting additional influence from nutritional, managerial, and environmental factors [71]. Acquired immunity and climatic and meteorologic factors may also alter relationships between parasite burden or diagnostic indicators and animal productivity. Exploring the interplay of these different factors using mathematical models, conducting fundamental research on the development of immunity against helminths [106], and validating insights in field studies may yield further progress in this area [107,108].

Alongside these recent insights, it also became clear that optimal decision-making requires greater inclusion of economic and social sciences to improve animal health and farmer uptake [109]. On the economic side, Van der Voort *et al.* showed that linking a farm’s technical production process with helminth infection levels and control strategies may open unexpected herd performance improvement pathways which are not necessarily related to the infection itself, even in highly infected herds [110]. Indeed, on economically inefficient farms – i.e., farms with a poor conversion rate from feed into milk – grazing management and anthelmintic interventions may not definitively increase outputs until those efficiency issues are solved [111]. Helminth control can thus be more beneficial on efficient farms with low-to-moderate infection levels than on inefficient farms with high infection levels [105,111], but this requires an appropriate economic analysis to define the farm’s production efficiency. On the socio-psychological side, farming objectives and farmer profiles and attitudes have a major impact on helminth control [15]. For instance, accounting for individual economic and environmental motives is critical when communicating helminth diagnostic and treatment strategies [114,115]. Risk perception or expressed concern about AR, on the other hand, may not or only weakly influence the intention to undertake time-consuming AR testing [116]. Both socio-psychology and practical hurdles or facilitators will greatly affect the actual helminth control strategy.

All the above factors determine how parasites impact farm performance. As long as these factors and their interplay are only partially understood, no decision algorithm can define an optimal farm-specific control strategy [117]. Veterinary advisors with (i) up-to-date knowledge of the tools to monitor and control helminth infections, their epidemiology and economic impact and (ii) the skills to collaborate with feed and economic advisors are thus crucial for evaluating the different factors in play on a given farm, and developing integrated evidence-based advice on helminth control.

# Concluding Remarks

Cattle production systems are facing increased pressure to raise outputs while contending with an increasingly unpredictable environment (environmental regulations, consumer expectations, climate change). Confronted with these challenges, treating only animals with clinical signs or preventatively treating entire herds may seem the simplest and least labour-intensive helminth control strategies. However, the production losses caused by subclinical cattle helminth infections and the growing threats of AR are rendering such strategies untenable. Alongside a strong international push for more sustainable agriculture methods [5,118], it is increasingly important to validate methods for detecting subclinical infections on farms. This will further the elimination of production losses, increase economic efficiency, and reduce total anthelmintic usage. We have described recent progress toward meeting these goals, including the development of new diagnostics for important cattle helminths and the definition of subclinical thresholds for existing tests. Universal thresholds that work across regions, production systems and farmer objectives may be difficult to achieve. Future studies are thus required to optimise these thresholds under a widening range of farm, climate, and grazing conditions as well as to incorporate new tests into the subclinical threshold paradigm.

Great strides have been made toward sustainable helminth control, yet much work remains to be done (see **Outstanding Questions**). Ongoing research developments must be linked to veterinary advice and the actual knowledge, attitudes, and practices of farmers themselves, ensuring that all relevant stakeholders have the up-to-date information necessary to make optimal anthelmintic treatment decisions.

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# Glossary

**Anthelmintic resistance (AR)**: the heritable genetic ability of a parasite to tolerate exposure to a standard anthelmintic drug treatment that would otherwise be lethal.

**Bulk tank milk (BTM)**: milk collected from bulk milk tanks, in which a dairy herd’s milk output is stored prior to transport to the processor.

**Faecal egg count (FEC)**: measurement of the number of nematode eggs shed in faeces, expressed in eggs per gram (EPG).

**First season grazing (FSG)**: describes calves entering their first season of pasture grazing, when natural immunity is low and the risk of helminth infection is particularly high.

**Pasture risk assessment**: estimating the risk of cattle helminth infections based on the animals’ grazing history, pasture management system, and anthelmintic treatment regimen.

***Refugia***:the proportion of the parasite population that is not exposed to a particular given control measure, thus escaping selection for resistance (e.g., eggs or larvae on pasture or parasitic stages in animals that are left untreated).

**Subclinical infection**: describes a helminth infection that does not induce clinical signs but may still impact production outputs (e.g., milk yield and/or weight gain). Detectable via diagnostic tests or from positive production responses after anthelmintic treatment.

**Subclinical threshold**: a quantitative cut-off (e.g., an FEC, a BTM ELISA measurement, an average daily weight gain level, etc.) indicating a subclinical parasitic infection that is likely to cause production losses.

**Targeted selective treatment (TST)**: anthelmintic treatment of individual animals within a group/herd based on one or several treatment indicators such as FEC, ELISA, weight gain, or diarrhoea score.

**Targeted treatment (TT)**:anthelmintic treatment of a whole herd or group of animals based on knowledge of infection risk or of parameters that quantify the severity of infection.

**Tolerance**: the ability of an animal to maintain health or performance under increasing parasite burden.

**Table 1**. **Summary of indicative production-based thresholds for cattle helminth diagnostics, derived from published literature or from proposed consensus interpretations.**

|  |  |  |  |
| --- | --- | --- | --- |
| **Helminth** | **Diagnostic test** | **Indicative thresholds** | **Refs** |
| *Ostertagia ostertagi* | Pepsinogen assay (serum/plasma) | Group mean of FSG animals ≥ 2.5 U Tyr: likely impact on productivity | [27,28] |
| SVANOVIR® *O.ostertagi*-Ab (serum) | Group mean of FSG animals ≥ 0.7 ODR: likely impact on productivity | [13,20,28] |
| Animal weight gain | Individual weight gain < group ADWG | [28,30] |
| Parasit’sim (simulation model) | ≥ 3 successive L3 generations on FSG animals’ pasture | [13] |
| SVANOVIR® *O.ostertagi*-Ab (BTM) | > 0.5 ODR: threshold for potential production losses≥ 0.8 ODR: threshold for potential significant production losses (milk yield reduced by ≥ 1 kg/cow/day) | [18,36,37,39,42] |
| SVANOVIR® *O.ostertagi*-Ab (individual milk) | ≥ 0.5 ODR: indicates exposure leading to potential production losses | [19,119–121] |
| TEC | < 8 months: high probability of production losses when exposed to significant larval challenge | [18,42] |
| *Fasciola hepatica* | Fluke burden | 1-10 flukes: threshold for effects on productivity>10 flukes: threshold for substantial effects on productivity | [66,70,71] |
| Liver fibrosis score | Mild focal fibrosis: some effects on productivity/weight gainSevere local or mild-to-severe generalised fibrosis: strong effects on productivity/weight gain | [70] |
| Faecal examination | Positive: threshold for effects on productivity; higher egg counts = increasing impacts | [70] |
| MonoscreenAgELISA – Bio-X Diagnostics (faeces) | Positive: threshold for effects on productivity, with higher S/P% = increasing impact | [60–63,66,70] |
| Various ELISA kits (serum and individual milk) | Positive (according to manufacturers’ thresholds): potential impact on productivity, with higher test results = likely increasing impact | [66,70] |
| SVANOVIR® *F.hepatica-*Ab (BTM) | ≥ 0.3 ODR: threshold for effects on productivity≥ 0.6 ODR: likely substantial effects on productivity | [73,77–79,83] |
| IDEXX® Fasciolosis Verification Test (BTM) | > 30 S/P%: threshold for effects on productivity> 80 S/P%: likely substantial effects on productivity | [74,122] |
| Monoscreen AbELISA – Bio-X Diagnostics (BTM) | ≥ 15 S/P%: threshold for effects on productivity≥ 45 S/P%: likely substantial effects on productivity | [80] |
| *Dictyocaulus viviparus* | Baermann technique (faeces) | Positive: likely impact on production | [87] |
| NL-ELISA (BTM) | Positive (≥ 10 ODR): likely impact on production | [101] |
| DE-ELISA (BTM) | 0.30 ≤ ODR < 0.41: positive with minimal impact on production≥ 0.41 ODR once, from regular testing during grazing season: threshold for effects on productivity≥ 0.41 ODR at least twice during grazing season: likely significant effects on productivity | [86,92] |

Abbreviations: DE-ELISA, *D. viviparus* ELISA developed by Hannover University, based on recombinant major sperm protein antigen; NL-ELISA, *D. viviparus* ELISA offered by the Animal Health Service (Royal GD, the Netherlands), based on purified low-molecular-weight product isolated from crude adult worm extracts; S/P%: sample-to-positive percentage.