Original Research Article

EPG and pasture larval count of gastrointestinal nematodes are strongly influenced by weather parameters: empirical evidence from Parbhani Marathwada

ABSTRACT

Aims: the present study was aimed to develop a bioclimatograph of gastrointestinal nematodes of cattle in Marathwada region of Maharashtra.

Study design: a standard protocol as per published reports was followed for recording of prevalence of GI nematodes from catte.

Place and Duration of Study: The study was conducted at livestock instructional farm of College of Veterinary and Animal Sciences Parbhani during the period February 2022 to January, 2023.

Methodology: In all total 253 faecal samples were collected from calves and adult cattle belonging to RK instructional farm, COVAS, Parbhani, of which 75 (30.00%) were positive. Prevalence of GI nematode infections was 38.84, 20.00 and 22.22% and EPG count as 90.09, 48.33 and 30.56% during monsoon, winter, and summer season, respectively. Sample collection and processing was done by following standard parasitological procedures and only fresh fecal droppings were used for the laboratory investigation.

Results: The study conducted on availability of larvae on pasture revealed the overall prevalence of 61 percent (87 pasture were positive out of 143 examined). The PLC analysis showed the highest distribution of larvae on pasture during winter season i.e. 57.81 %, followed by monsoon season i.e. 57.81 % and 00.0% during summer season. The Pasture larval count observed during different seasons was 23.83, 33.07 and 0.00 per 100 gm, respectively. The abstract information drawn from the bioclimatographs plotted for 2000-2021 and its validation with real time data of 2022 (Feb 2022-Jan 2023) was a) Suitable months for survival of *Heamonchus* and *Oesophagostomum* infective larvae on the pasture of the grazing land in year 2022-2023 were Jan - March and June – September and for *Trichostrongylus* Jan – March and October 2022- January 2023 and b)The climatic data and Bioclimatograph were plotted from year 2000-2021 and 2022+January 2023 showed the correlation between EPG and PLC.

Conclusion: The bioclimatographs can be developed and utilized according the regional agro-climatic variations and be useful for devising controlmeasures against gastrointestinal nematodes of livestock.

Keywords: bioclimatograph, cattle, nematodes, Maharashtra

1. INTRODUCTION

It is well known that the epidemiology of gastrointestinal parasites in livestock varies according to regional/local environmental circumstances, grazing resources/patterns, and management approaches. Accurate mapping of parasitic fauna of each livestock species from different agro-climatic seasons reveals vital information from which additional management methods can be derived [1]. Based on this fact present study also included the seasonal variation study of GI parasitism in cattle.

Assessment of infective larvae in pasture gives an idea of level of infection risk to animals grazed in a particular pasture land. Pasture herbage counts of infective larvae are used increasingly in the diagnosis and prognosis of parasitic disease in farm animals [2]. Similarly by using PLC and environmental data plotting of bio-climatographs suggest the best modelling strategies and making accurate predictions [3]. Similarly, contaminated pasture is the source of infection, so it is important to know the prevalence of infective larvae in the pasture for adopting suitable grazing practices and therefore management of pasture is an important component of nematode parasite control programs [4]. According to these two authors, it has also several benefits in relation to productivity including weight gain, improved feed conversion, increased milk production, better reproductive performance, greater carcass quality, improved immunological status, and reduced morbidity and mortality.

The main objective of choosing a parasite diagnostic technique is the reliability of the information towards solving the problem, however, due to ignorance or some or the other reasons, importance of free-living stages of GIN (eggs, developing larvae, and L3) is not generally taken into account for forming effective technique of parasitic diagnosis [5]. Under such scenario, one of the best methods is the pasture larval count (PLC), which attempts to quantitatively and qualitatively identify the nematode larval species to support research on the dynamics of the parasite population. The PLC can also spot monthly and seasonal variations in pasture infectivity, giving grazing animals a risk score for parasite exposure [6,7]. In view of this, the current study was planned for estmation of Pasture larval burden and its correlation with the climatic factors at Parbhani area.

2. MATERIAL AND METHODS

Present research work was undertaken with the objectives to study the gastrointestinal nematodes infecting to livestock at Parbhani region. The research work was undertaken at Department of Veterinary Parasitology, College of Veterinary and Animal Sciences, (MAFSU) Parbhani. Parbhani city is situated at 190 23' North latitude 760 79' East longitudes and at an altitude of 454 m above mean sea level in Marathwada division of Maharashtra state. Parbhani tract comes in rain shadow area receiving more than 80 per cent of the rainfall from south-west monsoon. Agro climatically this area falls in assured rainfall zone of Maharashtra state. The mean average annual precipitation about 938.7 mm, mostly received between June to September from south western monsoon. The winter rains are uncertain and scanty. The mean maximum temperature varies from 28.7-42.1oC (33.7oC), whereas the mean minimum temperature varies from 9.0oC to 26.5oC(18.5oC) during winter and summer respectively. Thus, Parbhani has cold winter and dry hot summer. The Parbhani experiences Relative Humidity, RH-I 41-87%(71%) and RH-II(98%). Southwest monsoon season-June to September, Post monsoon season-October to January and summer season- February to May [8].

The research was carried out from February 2022-January 2023. The host animals included in the study were Red Kandhari Cattle of Marathwada region. The animals were selected from Red Kandhari Research and Instructional Farm of COVAS Parbhani.

Following seasons were considered for the present study. Seasons were as per months and weeks of a calendar according to standard meteorological norms designed for Marathwada region (VNMKV Vision- 2020, 1998). Seasons defined as per World Meteorological Organization(WMO) norms for the Marathwada region are monsoon, winter and summer.

Monsoon: 4th June- 4th November (23rd -44th week)

Winter : 5th November-4th March (45th -9th week)

Summer : 5th March-3rd June (10th -22nd week)

Six adult cattle and six calves were selected randomly from the ILFC, College of Veterinary and Animal Sciences, Parbhani. These animals are not dewormed throughout experimental period. However, they were treated for other illness and health fitness. EPG of the faecal samples from the selected animals were estimated twice in a month by Stoll's Egg counting technique as described earlier [2]. Pasture larval count (PLC) were estimated twice or thrice once in a month throughout experimental period by following standard procedure [9]. The recovered larvae from the herbage samples were killed and stained with Lugol's iodine solution for few minutes . Then a drop containing two-three larvae was taken on a glass slide with the help of pasture pipette and covered with a coverslip. It was examined under a compound microscope fixed with an occulomicrometer in the eyepiece. The Total Length of the larvae (TL) and Sheath Tail Extension (STE) was measured by occulomicrometer. The readings were subsequently calculated and converted into micrometres. The obtained lengths of TL and STE of larvae were matched according to the standard measurements of the infective larvae of gastrointestinal nematodes of sheep [10]. The data obtained from various parameters was analyzed by employing simple correlation, multiple regression and completely randomized design using computer application, WASP version 2.0 (www.ccari.res.in)

3. RESULTS AND DISCUSSION

3.1 Prevalence of Gastro intestinal nematodes in cattle at Parbhani region

In all total 253 faecal samples were collected from calves and adult cattle belonging to RK instructional farm, College of Veterinary and Animal Sciences, Parbhani. Out of 253 samples examined 75 (30.00%) were positive for different species of GI nematode infection.

At par prevalence was reported from Uttar Pradesh [11], from Gujrat [1] and from Meghalaya [12] states of India. Few scientists reported very less prevalence of GI parasitism 12.50 % [13]; whereas very high prevalence of 75% by some of the researchers [14]. Comparison of prevalence from one geographic region with other region is not justifiable because

1. The development, growth, survival and transmission of infective larval stages on pasture is greatly influenced by rainfall, temperature, humidity, soil moisture and

other conditions of a particular region. All these vary from place to place, country to country and year to year.

2. Similarly prevalence also depends on host factors such as breed and animal husbandry practices, which all differ at different locations.

During three different seasons the prevalence of GI nematode infections was 38.84%, during monsoon, 20.00% during winter and 22.22% during summer season.Study recorded the more incidences of Strongyles and Strongyloides spp. It is well known fact thatduring rainy seasonthe survival of infective stages on pasture is for longer period of time,whichfacilitatesuptake of infective stages by host and it results in increased prevalence. During winter season due to presence of infective stages on pasture nearly same or to certain extent lesser infection occur. In summer season, it is devoid of optimumgeoclimaticconditionsi.e higher temperature and relative humidity and minimum level of moisture are available, as a result nil or lowest level of infection occur.Seasonal prevalence for GI parasitism is also reported by several reserchers [11, 13, 15].

Table 1 Prevalence and Eggs Per Gram (EPG) for nematode infections in Cattle at Parbhani region.

Season	**Prevalence of GI parasitism**		**%**	**EPGvalues**		
	(TE)	(TP)		No of observations	Mean ± SE	Range

Monsoon	121	47	38.84	47	90.09 a± 11.934	0-500
Winter	60	12	20.00	12	48.33b±13.96	0-400
Summer	72	16	22.22	16	30.56b±8.07	0-400
	253	75	30.00		HS	
Stat				CD Value	CD(0.01) = 47.735	
					CD(0.05) = 36.320	

TP: Total Positive; TE: Total Examined: HS: Highly Significant

The mean EPG count observed during monsoon season was highest as 90.09, moderate during winter season as 48.33 and lowest as 30.56 during summer season. Different workers has reported the EPG from the country as detailed in table(.3).

3.2 Prevalence of Pasture larvae of nematodes of cattle at Parbhani region

The study conducted on availability of larvae on pasture revealed the overall prevalence of 61 percent (87 pasture samples were positive out of 143 examined). The PLC analysis showed the highest and equal distribution of larvae on pasture during winter season and monsoon season i.e. 57.81 per cent. Negligible or no larvae were observed on pasture during summer season.

From India the PLC levels were reported from Assam [16] and from Sikkim [17].Both the authors reported PLC throughout the year with seasonal variation. However, in the present study in Maharashtra PLC levels were only observed during monsoon and winter season. Such variation in the PLC levels might be attributed to the difference in geographic location. In the present study conducted during the year 2022 there was a highest rainfall and hence maximum number of larvae were available on pasturewhich enhanced the chances of infection in animals during monsoon followed by winter [18]. Furthermore the monsoon onset occurred in July and ended in September with occasional rains up to December, with the highest rainfall in August. This would have made the environmental conditions more favourable for the development and survival of the pre-parasitic stages leading to increased availability of infective larvae on the pasture during monsoon and subsequent months [19].

Table 2 Prevalence of Pasture larval count (PLC) for nematodes of cattle at Parbhani region

Season	**Prevalence of GI parasitism**			**PLC values**		
	TE	**TP**	**%**	**No of observations**	**Mean ± SE**	**Range**
Monsoon	64	37	57.81	37	23.83a± 3.575	0-105
Winter	57	50	87.71	50	33.07a± 4.034	0-83
Summer	22	0	0.00	0	0.00b	0.00
	143	87	61.00		S	
				CD Value	CD(0.01) = 21.115	
					CD(0.05) = 16.066	

TP:Total Positive; TE:Total Examined: S: Significant

<u>3.3 Pasture Larval Count (PLC)</u>

The Pasture larval count observed during different seasons was 23.83, 33.07 and 0.00 per 100 gm of pasture during monsoon, winter and summer season, respectively. PLC reported by various workers is abstracted in table (5). Except a single study from Sikkim [17]; no much work from India is available for comparison and discussion.

3.4 Identification of the larvae of helminth parasites recovered from the pasture

In the present study various species of gastrointestinal nematodes found during the morphometric examination of larvae. The larvae recovered from the pasture were identified as *Bunostomumphlebotomum*, *Bunostomumspp*, *Haemonchuscontortus*, *Trichostrongylus spp*., *Oesophagostomumradiatum*, *Cooperia punctata*, *Ostertegiaostertegi* and *Strongyloidespapillosus* based on morphometric observations and its equating it with the standard morphometric observations (Table – 6).

Among various species *Oesophagostomumradiatum*was the most prevalent as compared to other followed by *Bunostomumphlebotomum*and *Haemonchuscontortus*etc. Mixed infection other than GI nematodes was also encountered such as *Schistosoma* infection in calves.

3.5 EPG and its correlation with weather parameters

The EPG levels was found to be significantly correlated with RH (E), while, with other parameters like Tmin, Tmax, BSS, EVP, TRF are negatively non-significantly correlated and RH-M positively non-significantly correlated. In nutshell humidity levels are positively correlated while rests of the parameters are having negative (inverse) relationship. Positive correlation of RH factor indicates that in the tropical region like Parbhani where abundant quantum of heat (temperature) is available, humidity level matters.

3.6 PLC and its correlation with weather parameters

As like EPG correlations, more or less similar pattern of correlations between PLC levels and weather parameters was observed. The only difference noted was that, RH-E has also shown negative correlation, though its magnitude is non-significant. It indicates that in the region where grazing of animals is done on pasture, humidity levels in the morning has more impact. It helps in crawling and transfer of larvae on pasture/grass blades.

EPG levels and PLC levels showed negative correlation with each other.

Table 3 Showing correlation matrix for EPG, PLC and weather parameters of Parbhani region.

Correlation Matrix

-	T_{min}(°C)	T_{max}(°C)	RH-M(%)	RH-E(%)	BSS(Hrs.)	EVP(mm)	TRF(mm)	EPG	PLC
T_{min}(°C)	1.000								
T_{max}(°C)	0.116	1.000							
RH-M(%)	-0.890	-0.070	1.000						

RH-E(%)	-0.662	0.374	0.656	1.000					
BSS(Hrs.)	-0.468	0.486	0.521	0.622	1.000				
EVP(mm)	-0.473	0.477	0.526	0.615	1.000	1.000			
TRF(mm)	0.938	0.191	-0.926	-0.640	-0.515	-0.520	1.000		
EPG	-0.544	-0.087	0.435	0.705	-0.033	-0.041	-0.457	1.000	
PLC	-0.374	-0.386	0.443	-0.002	-0.128	-0.120	-0.475	-0.008	1.000

Table 4 Showing Student T-test for EPG, PLC and weather parameters of Parbhani region.

Variables Tested	T Value	T Table	Significance at 5%
Tmax -RH(M)	5.865	2.262	Significant
Tmax -RH(E)	2.651	2.262	Significant
Tmax -EVP	8.097	2.262	Significant
RH(M) -RH(E)	2.605	2.262	Significant

RH(M) -EVP	7.36	2.262	Significant
RH(E) -TRF	2.384	2.262	Significant
RH(E) -BSS	2.34	2.262	Significant
RH(E) -EVP	2.499	2.262	Significant
RH(E) -EPG	**2.98**	**2.262**	**Significant**
TRF -BSS	190.376	2.262	Significant
Tmax -Tmin	0.349	2.262	Non Significant
Tmax -TRF	1.591	2.262	Non Significant
Tmax -BSS	1.61	2.262	Non Significant
Tmax -EPG	1.945	2.262	Non Significant
Tmax -PLC	1.209	2.262	Non Significant
Tmin -RH(M)	0.211	2.262	Non Significant
Tmin -RH(E)	1.211	2.262	Non Significant
Tmin -TRF	1.666	2.262	Non Significant
Tmin -BSS	1.63	2.262	Non Significant

Tmin -EVP	0.583	2.262	Non Significant
Tmin -EPG	**0.262**	**2.262**	**Non Significant**
Tmin -PLC	**1.257**	**2.262**	**Non Significant**
RH(M) -TRF	1.832	2.262	Non Significant
RH(M) -BSS	1.857	2.262	Non Significant
RH(M) -EPG	**1.45**	**2.262**	**Non Significant**
RH(M) -PLC	**1.481**	**2.262**	**Non Significant**
RH(E) -PLC	**0.005**	**2.262**	**Non Significant**
TRF -EVP	1.802	2.262	Non Significant
TRF -EPG	**0.1**	**2.262**	**Non Significant**
TRF -PLC	**0.389**	**2.262**	**Non Significant**
BSS -EVP	1.829	2.262	Non Significant
BSS -EPG	**0.122**	**2.262**	**Non Significant**

Regression analysis value R^2(square)=82.95%indicated the role of environmental factors to the extent of 82.9% and remaining almost 17% remains unexplained or factors could not be predicted. The regression analysis indicates RH(E), TRF,BSS,EVP are negatively correlated and showing negative impact on EPG count while Tmax, Tmin ,RH(M) are positively correlated having positive impact on EPG levels.

Table 5 Regression analysis – EPG as dependent factor and all environmental factors and PLC as independent factors

Independent Variables	Average	Reg. coefficients (b)	Standard Error(SE(b))	T Test	T table (0.05)
T_{min}(℃)	32.936	0.530	nan	nan	4.303
T_{max}(℃)	19.291	0.065	nan	nan	4.303
RH-(M) (%)	19.291	0.000	nan	nan	4.303
RH-(E) (%)	78.818	-39.894	nan	nan	4.303
BSS(Hrs.)	69.636	-38.047	nan	nan	4.303
EVP (mm)	98.064	-521.110	nan	nan	4.303
TRF (mm)	98.727	-522.226	nan	nan	4.303
PLC	74.996	93.514	nan	nan	4.303

Intercept (a) = 148.171

Coefficient of determination (R Square) = 82.95 %

Multiple Correlation Coefficient (R) = 0.977

Standard Error = 26.541

The resultant equation for regression model derived as –

EPG = 148.171+(-4.073) **x** Tmin+(-2.647) **x** Tmax+(0.479) **x** RH-M+(1.759) **x** RH-E+(6.915) **x** TRF+(-7.204) **x** BSS+(-0.958) **x** EVP+(-0.251) **x** PLC+26.541

In another set of anslysis PLC was taken as a dependent factor and weather parameters and EPG were taken as independent factors. here the regression analysis value *i.e* R^2(square)=88.4 % indicates the environmental factors has played role to the extent of 88.4 % and remaining almost 11.6 % remains unexplained or could not be predicted. The regression analysis also indicated that RH-M and BSS are negatively correlated and shows negative impact on PLC count while Tmax, Tmin, RH(E), EVP and TRF are positively correlated with PLC and has got positive impact on PLC levels.

Table 6 Regression analysis –PLC as dependent factor and all environmental factors and EPG as independent factors

Independent Variables	Average	Reg. coefficients (b)	Standard Error(SE(b))	T Test	T table (0.05)
T_{min}(℃)	32.936	13.282	12.329	1.077	4.303
T_{max}(℃)	19.291	0.651	6.429	0.101	4.303

RH-(M) (%)	78.818	-6.239	5.005	-1.246	4.303
RH-(E) (%)	69.636	12.671	5.667	2.236	4.303
BSS(Hrs.)	98.064	-86.766	50.763	-1.709	4.303
EVP(mm)	98.727	86.369	50.861	1.698	4.303
TRF(mm)	6.565	2.325	15.942	0.146	4.303
PLC	74.996	-0.561	0.979	-0.573	4.303

Intercept (a) = -797.320

Coefficient of determination (R Square) = 88.4 %

Multiple Correlation Coefficient (R) = 0.940

Standard Error = 39.662

The resultant equation for regression model of PLC is derived as –

PLC = -797.320 +(13.282) **x** Tmin+(0.651) **x** Tmax+(-6.239) **x** RH-M+(12.671) **x** RH-E+(-86.766) **x** TRF+(86.369) **x** BSS+(2.325) **x** EVP+(-0.561) **x** EPG+39.662

It is evident from the above regression model that PLC has positive impact with EPG, meaning if PLC rises ECG count also rises. It is also not surprising that TRF showing negative impact and the reason could be high rainfall during the year 2022 might be responsible for washing and run-off of the larvae from the pasture and reduction in infection due to less intake of grass from pasture land due to high water content in pasture land.

It is well known fact that weather and climate at the particular geographic location plays crucial role in build-up of population of helminthic infections in ruminants [3]. The climatic factors such as temperature, precipitation, humidity, wind direction, velocity, sun light, bright sunshine hours, are the real determinants of helminthic infections. Workers from different parts of the world through their research proved that, for development, survival of parasitic stages of strongyle nematodes of ruminants and their transmission and availability on pasture are governed by temperature and moisture [20, 21].

After hatching from parasite eggs, the first stage larvae (L.) feed on bacteria in faeces, moult to second stage which also feeds on bacteria and then moult to ensheathed infective third stage larvae (L3). All these life cycle changes happen in the nature on grazing land with the availability of temperature, atmospheric oxygen and adequate moisture. Thus in particular tropical parts, rainfall happens to be the greatest determinant in deciding the life cycle of helminth parasites. The L3 stage larvae migrate out of the faeces and disseminate onto the vegetation and remains there until a potential host ingests them or until they die. However, such exogenous phase of the life cycle consists of two independent processes such as (i) development of infective larvae, and (ii) transmission of infective larvae on the grass blades to increase the likely chances of infection to definitive host [3]. The environmental conditions which favour, one may not favour the other. The optimum temperature for development is generally higher than the optimum temperature for survival and transmission. The grazing system together with the rate of development of larvae can be used as essential information to predict the chances of infection, their peak period and hence such information can be valuable for forming deworming schedule.

4. CONCLUSION

From results of the current study, it is concluded that the suitable months for survival of *Heamonchus* and *Oesophagostomum* infective larvae on the pasture of the grazing land in year 2022-2023 were Jan - March and June – September and for *Trichostrongylus* Jan – March and October 2022- January 2023. Therefore the deworming schedule should be formulated in this region accordingly.

REFERENCES

References must be listed at the end of the manuscript and numbered in the order that they appear in the text. Every reference referred in the text must also present in the reference list and vice versa. In the text, citations should be indicated by the reference number in brackets [3].

1. Maharana, B. R., B. Kumar, N. R. Sudhakar, S. K. Behera, & T. K. Patbandha. Prevalence of gastrointestinal parasites in bovines in and around Junagadh (Gujarat). Journal of parasitic Diseases. 2016; 40(4): 1174-1178.

2. Soulsby, E.J.L. Helminth,Arthropod&Protozoa of domesticated Animals. London: Baillière Tindall, 1965; 7th Edition
3. Van Dijk, J., N.D. Sargison., F. Kenyon., and P.J. Skuce. Climate change and infectious disease: helminthological challenges to farmed ruminants in temperate regions. Animal. 2010; 4(3): 377-392.
4. Stromberg, B. E. Environmental factors influencing transmission. Veterinary Parasitology. 1997; 72(3-4): 247-264
5. Stromberg, B. E., and G. A. Averbeck,. The role of parasite epidemiology in the management of grazing cattle. International Journal for Parasitology. 1999; 29(1): 33-39.
6. Almería, S., and J. Uriarte,. Dynamics of pasture contamination by gastrointestinal nematodes of cattle under extensive management systems: proposal for strategic control. Veterinary Parasitology, 1999; 83(1): 37-47.
7. Dakhore, K.D., E.K. Yadav and A.R. Shaikh. In Climatic Normals and Extreme Events at Parbhani. Published by Dr D P Waskar, Director of Research, Vasantrao Naik Marathwada Krishi Vidyapeeth, Parbhani 431402 (M.S.) 2020; VNMKV/DOR/Technical Bulletin/10/2020 pp01-02.
8. Martin, R. R., I. Beveridge, A. L. Pullman., and T. H. Brown. A modified technique for the estimation of the number of infective nematode larvae present on pasture, and its application in the field under South Australian conditions. Veterinary parasitology, 1990; 37(2): 133-143.

9. Van Wyk, J. A., and E. Mayhew,. Morphological identification of parasitic nematode infective larvae of small ruminants and cattle: A practical lab guide. Onderstepoort Journal of Veterinary Research. 2013; 80(1): 1-14.

10. Singh, A., A. K. Gangwar., N. K. Shinde., and S. Srivastava. Gastrointestinal parasitism in bovines of Faizabad. Journal of Veterinary Parasitology. 2008; 22(1): 31-33.

11. Laha, R., M. Das, and A. Goswami, (). Gastrointestinal parasitic infections in organized cattle farms of Meghalaya. Veterinary World. 2013; 6(2): 109

12. Wadhwa, A., R. K. Tanwar., L. D.Singla., S. Eda., N.Kumar., and Y. Kumar. Prevalence of gastrointestinal helminthes in cattle and buffaloes in Bikaner, Rajasthan, India. Veterinary World. 2011; 4(9): 417.

13. Marskole, P., Y. Verma., A. K Dixit., and M. Swamy. Prevalence and burden of gastrointestinal parasites in cattle and buffaloes in Jabalpur, India. Veterinary World. 2016; 9(11): 1214.

14. Raman, M. A. S. S. Pandian and I. Manikkavasagan. Impact of climatological parameters on prevalence of gastrointestinal helminths of small ruminants in Tamil Nadu. Journal of Agrometeorology. 2015; 17(2): 256- 258.

15. Das, M., D. K. Deka., S. Islam., P. C. Sarmah., and K. Bhattacharjee. Gastrointestinal nematode larvae in the grazing land of cattle in Guwahati, Assam. Veterinary World. 2016; 9(12): 1343.

16. Pal, P., L.R. Chatlod., and R.K. Avasthe. Preparation of bioclimatograph for Haemonchosis and trichostrongylosis in goats of subtropical high humid zone of Sikkim. The Indian Journal of Animal Sciences, 2017; 87(7).
17. Durie, P. H. Parasitic gastro-enteritis of cattle: The distribution and survival of infective strongyle on pasture. Australian Journal of Agricultural Research. 1961; 12(6): 1200-1211.
18. Jithendran, K. P., and T. K. Bhat. Epidemiology of parasitoses in dairy animals in the North West Humid Himalayan Region of India with particular reference to gastrointestinal nematodes. Tropical Animal Health and Production. 1999; 31(4): 205-214.
19. Swarnkar, C.P. and Singh, D. Influence of annual rainfall on epidemiology of gastrointestinal parasites in sheep flocks of Rajasthan. Indian Journal of Animal Sciences. 2014; 84(11): 1171-1176.
20. Melville LA, Van Dijk J, Mitchell S, Innocent G, Bartley DJ. Variation in hatching responses of Nematodirusbattus eggs to temperature experiences. Parasites and Vectors. 2020; 13(1): 494. https://doi.org/10.1186/s13071-020-04368-9

Marcelo Beltrão Molento, Andréia Buzatti, Lew Kan Sprenger,. Pasture larval count as a supporting method for parasite epidemiology, population dynamic and control in ruminants. Livestock Science; 2016; 192: 48-54. https://doi.org/10.1016/j.livsci.2016.08.013