Systematic Review

Epidemiology of Ticks and Tick-Borne Pathogens in Domestic Ruminants across Southern African Development Community (SADC) Region from 1980 until 2021: A Systematic Review and Meta-Analysis

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Abstract: Ticks are hematophagous ectoparasites that are capable of infesting a wide range of mammals, including domestic animals, ruminants, wildlife, and humans across the world, and they transmit disease-causing pathogens. Numerous individual epidemiological studies have been conducted on the distribution and prevalence of ticks and tick-borne diseases (TBDs) in the Southern African Developing Community (SADC) region, but no effort has been undertaken to synchronize findings, which would be helpful in the implementation of consolidated tick control measures. With the aim of generating consolidated pooled prevalence estimates of ticks and TBDs in the SADC region, we performed a systematic review and meta-analysis of published articles using the PRISMA 2020 guidelines. A deep search was performed on five electronic databases, namely, PubMed, ScienceDirect, Google Scholar, AJOL, and Springer Link. Of the 347 articles identified, only 61 of the articles were eligible for inclusion. In total, 18,355 tick specimens were collected, belonging to the genera *Amblyomma*, *Haemaphysalis*, *Hyalomma*, and *Rhipicephalus* (including *Boophilus*) across several countries, including South Africa (n = 8), Tanzania (n = 3), Zambia (n = 2), Zimbabwe (n = 2), Madagascar (n = 2), Angola (n = 2), Mozambique (n = 1), and Comoros (n = 1). The overall pooled prevalence estimate (PPE) of TBPs in livestock was 52.2%, with the highest PPE in cattle [51.2%], followed by sheep [45.4%], and goats [29.9%]. For bacteria-like and rickettsial TBPs, *Anaplasma marginale* had the highest PPE of 45.9%, followed by *A. centrale* [14.7%], *A. phagocytophilum* [2.52%], and *A. bovis* [0.88%], whilst *Ehrlichia ruminantium* had a PPE of 4.2%. For piroplasmids, *Babesia bigemina* and *B. bovis* had PPEs of 20.8% and 20.3%, respectively. *Theileria velifera* had the highest PPE of 43.0%, followed by *T. mutans* [29.1%], *T. parva* [25.0%], and other *Theileria* spp. [14.06%]. Findings from this study suggest the need for a consolidated scientific approach in the investigation of ticks, TBPs, and TBDs in the whole SADC region, as most of the TBDs are transboundary and require a regional control strategy.

Keywords: SADC; ticks and tick-borne pathogens; *Amblyomma*; *Boophilus*; *Haemaphysalis*; *Hyalomma*; *Rhipicephalus*; *Anaplasma*; *Ricketssia*; *Babesia*; *Theileria*

1. Introduction

Ticks are a major group of the hematophagous arthropods that are a veterinary and public health concern, resulting in major financial suffering for the agricultural sector [1]. Primarily, ticks are normally pathogen transmitters and are found in bushes, forests, and semi-arid areas [2]. The availability and behaviour of ticks highly depend on the climatic

conditions of a geographical area [3–5]. Subsequently, the alteration of the climatic conditions of an area can lead to a high prevalence of ticks and tick-borne disease, as observed in Tunisia [6].

The Southern African Developing Community (SADC) region has tick species that belong to three families, including *Argasidae*, *Ixodidae*, and *Nuttalliellidae* [7–11]. *Ixodidae*, which are known as hard ticks, are blood-sucking arthropods of amphibians, avians, reptiles, and mammals and they include various genera, such as *Ixodes*, *Amblyomma*, *Dermacentor*, *Haemaphysalis*, *Hyalomma*, and *Rhipicephalus* [12]. *Argasidae* is a family of soft ticks that does not lack scutum, as compared to the hard tick family; it has four genera, including *Argas*, *Carios*, *Ornithodoros*, and *Otobius* [10]. Lastly, the family *Nuttalliellidae* has only one tick species known as *Nuttalliella namaqua*, which shares similar characteristics to *Argasidae* and *Ixodidae* ticks [13,14].

Generally, adult ticks target medium- and large-sized hosts (animals and sometimes humans) for blood meal, whilst larvae and nymphs target small hosts for their survival [15]. These ticks are known to be global vectors of microorganisms infecting both animals and humans, such as bacteria, helminths, protozoal, ricketssial, and viral pathogens [16,17].

Ticks are distributors of tick-borne diseases (TBDs) amongst tropical and subtropical regions of the world [18–20]. Based on epidemiology, tick-borne diseases vary according to unequally shared space and time in various nations that depend specifically on the availability of certain tick species [21]. In urban and suburban areas, small rodents, hedgehogs, and shrews are well-known reservoirs of *Ixodes* ticks species, which transmit tick-borne pathogens [TBPs] [22]. This suggests that ticks are available in suburban green areas and the diversity and rate of tick-borne diseases in those areas might be equivalent to rural areas when quantified [23]. The TBPs may be more accurately detected by the use of more sensitive molecular techniques, such as conventional polymerase chain reaction (PCR), nested PCR (nPCR), real-time polymerase chain reaction (qPCR), reverse transcription-polymerase chain reaction (RT-PCR), and reverse line blot hybridization (RLB) [24,25]. Numerous TBPs, including *Anaplasma* spp., *Babesia* spp., *Ehrlichia* spp., *Ricketssia* spp., and *Theileria* spp., have been documented as significant pathogens of domestic ruminants, including cattle, goats, and sheep, and are further transmitted by ticks within individual SADC region countries [26–29].

For effective control measures against ticks and TBPs to be successful, it is pertinent that the distribution of ticks and TBPs within that particular geographical area is known. Therefore, this study was undertaken to generate consolidated data of tick abundance, as well as TBPs' prevalence and distribution, from blood and feeding ticks collected from domestic ruminants in the SADC region, using systematic review and meta-analysis.

2. Results

2.1. Literature Search and Eligible Studies

A total search of 33,494 articles was initially identified through PubMed (n = 56), Science Direct (n = 751), Google Scholar (n = 31,700), AJOL (n = 244), and Springer Link (n = 743). After the removal of duplicates, 32,097 articles remained. Of these, 31,929 articles were excluded after assessing the titles and abstracts. The remaining 168 full articles were then assessed for eligibility, and 107 from the following groups were excluded: (i) studies on non-domestic ruminants (n = 38); (ii) studies with insufficient data analysis (n = 46); and (iii) experimental studies (n = 23). Finally, 61 articles that reported on the prevalence of ticks and tick-borne pathogens detected from blood and tick samples of ruminants across the SADC region fulfilled the criteria for inclusion (Figure 1).
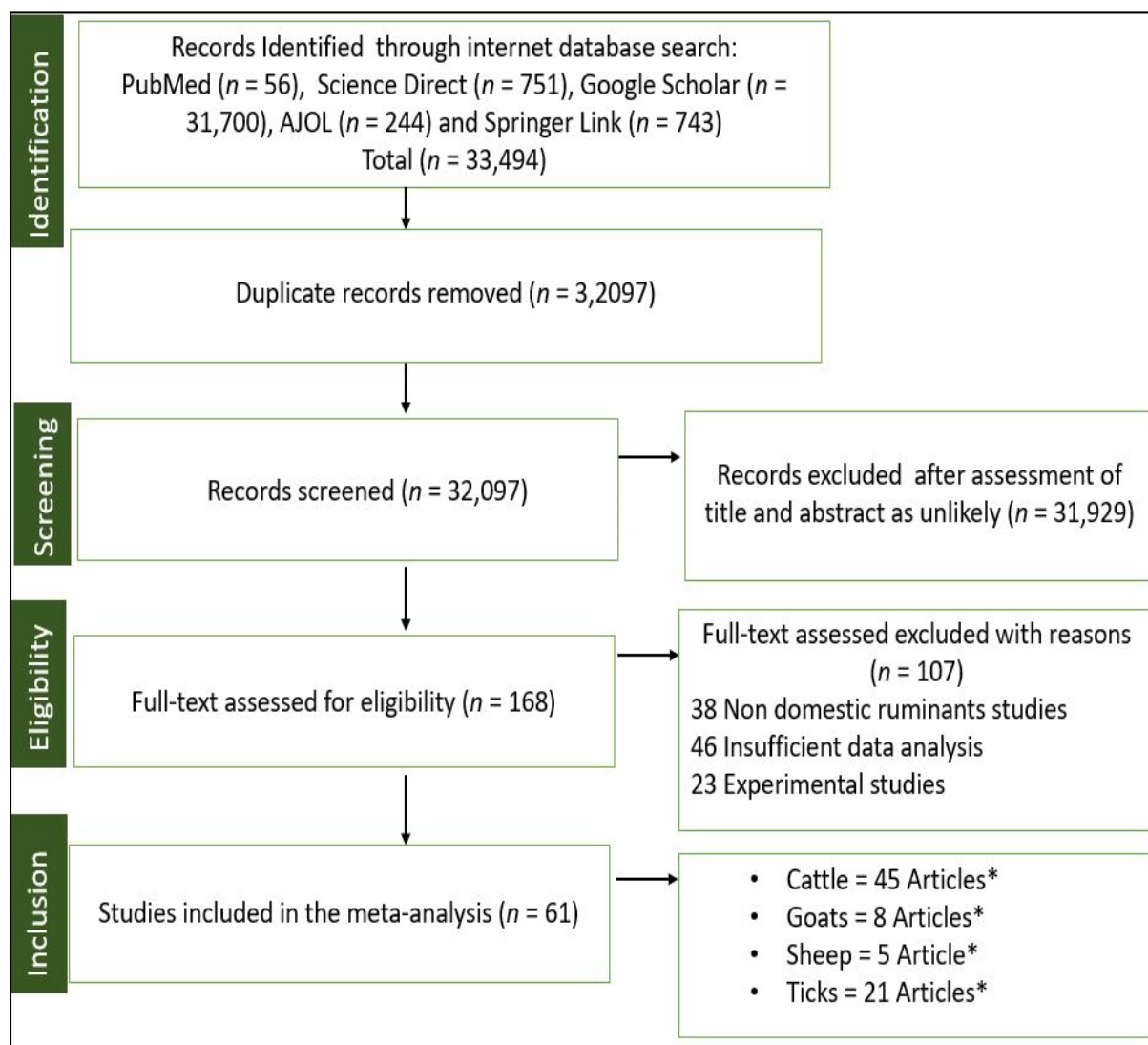
Figure 1. Flow chart of included studies, according to Preferred Reporting Items for Systematic Reviews and Meta-Analyses guidelines. * Whilst n = 61 articles were used for the meta-analysis, in some cases, the same article has published data from different ruminants hosts as well as ticks; hence, the total number of articles may appear as if it is more than 61.

2.2. Characteristics of Eligible Studies

The occurrence of tick-borne pathogens was detected in three livestock ruminant groups, namely, cattle (n = 45), goats (n = 8), and sheep (n = 5), with a total of 12,693 examined blood samples, published in any of the SADC countries (Table 1). These studies were conducted in South Africa (n = 18); Tanzania (n = 10); Zambia (n = 7); Mozambique (n = 5); Angola (n = 4); Botswana (n = 2); and Zimbabwe (n = 1). The blood sample size ranged between 8 and 960 per study and the individual prevalence of tick-borne pathogens in different animal species ranged from 0.33% to 100% (Table 1). Additionally, 21 studies focused on the prevalence of TBPs in tick species, which were conducted in South Africa (n = 8), followed by Tanzania (n = 3), Zambia (n = 2), Zimbabwe (n = 2), Madagascar (n = 2), Angola (n = 2), Mozambique (n = 1), and Comoros (n = 1). The tick sample size ranged between 36 and 7364 per study and the individual prevalence of TBPs in different tick species ranged from 0.02% to 62.32% (Table 2).

Table 1. Characteristics of all eligible studies reporting the occurrence of tick-borne pathogens in domestic ruminants across the Southern African Developing Community region.

Countries	Hosts	Sample Size	Total No. of Pathogens Detected	Pathogens Detected (No. of Positives, Prevalence (%)	Reference
Angola	Cattle	98	11	*A. platys* (n = 3; 3.06%), *A. capra* (n = 6; 6.12%), *A. phagocytophilum* (n = 2; 2.04%)	[30]
Angola	Cattle	76	51	*A. marginale* (n = 29; 38.15%), *Anaplasma* spp. (n = 6; 7.89%), *B. bigemina* (n = 2; 2.63%), *T. velifera* (n = 22; 28.95%), *Theileria* spp. (n = 6; 7.89%)	[31]
Angola	Goats	13	13	*A. ovis* (n = 13; 100.00%)	[30]
Angola	Cattle	88	78	*A. bovis* (n = 1; 1.14%), *A. centrale* (n = 11; 12.50%), *A. marginale* (n = 25; 28.41%), *Anaplasma* spp. (n = 22; 25.00%), *Anaplasma* spp (n = 22; 25.00%), *A. platys* (n = 16; 18.18%), *B. bigemina* (n = 35; 39.77%), *B. rossi* (n = 1; 1.14%), *E. ruminatium* (n = 3; 3.41%), *T. velifera* (n = 69; 78.41%), *T. mutans* (n = 65; 73.86), *Theileria* spp. (n = 63; 71.59%)	[32]
Angola	Goats	82	2	*A. centrale* (n = 2; 2.44%)	[32]
Angola	Sheep	85	68	*A. centrale* (n = 2; 2.35%), *A. marginale* (n = 1; 1.18%), *Anaplasma* spp. (n = 4; 4.71%), *A. platys* (n = 5; 5.88%), *B. bovis* (n = 1; 1.18%), *T. ovis* (n = 68; 80.00%), *Theileria* spp. (n = 46; 54.12%)	[32]
Angola	Cattle	76	38	*B. bigemina* (n = 38; 50.00%)	[33]
Botswana	Cattle	276	2	*T. mutans* (n = 1; 0.36%), *T. taurotragi* (n = 1; 0.36%)	[34]
Botswana	Goats	100	76	*A. ovis* (n = 76; 76.00%)	[35]
Botswana	Cattle	429	135	*Anaplasma* spp. (n = 135; 31.47%)	[36]
Malawi	Goats	99	74	*A. ovis* (n = 61; 61.62%), *Anaplasma* spp. (n = 74; 74.75%)	[37]
Malawi	Sheep	8	8	*A. ovis* (n = 8; 100%), *Anaplasma* spp. (n = 8; 100%)	[37]
Mozambique	Cattle	219	213	*A. marginale* (n = 213; 97.26%), *A. phagocytophilum* (n = 6; 2.74%), *Anaplasma* spp. (n = 191; 87.21%)	[38]
Mozambique	Cattle	477	323	*A. centrale* (n = 20; 4.19%), *A. bovis* (n = 4; 0.84%), *A. marginale* (n = 42; 8.80%), *B. bigemina* (n = 267; 55.97%), *B. bovis* (n = 201; 42.14%), *Ehrlichia* spp. (n = 29; 6.08%), *T. mutans* (n = 250; 52.41%), *T. taurotragi* (n = 5; 1.05%), *T. velifera* (n = 255; 53.46%), *Theileria* spp. (n = 41; 8.59%)	[39]
Mozambique	Cattle	117	104	*B. bigemina* (n = 104; 88.89%), *B. bovis* (n = 97; 82.91%)	[40]
Mozambique	Cattle	210	31	*E. ruminatium* (n = 31; 14.76%)	[41]
Mozambique	Cattle	49	6	*B. bigemina* (n = 6; 12.24%)	[33]
South Africa	Cattle	66	51	*A. centrale* (n = 27; 40.91%), *A. marginale* (n = 51; 77.27%)	[42]
South Africa	Cattle	517	295	*A. centrale* (n = 88; 17.02%), *A. marginale* (n = 295; 57.06%)	[43]
South Africa	Cattle	200	54	*T. parva* (n = 54; 27.00%)	[44]
South Africa	Cattle	149	88	*A. marginale* (n = 88; 59.06%)	[45]
South Africa	Cattle	846	140	*T. parva* (n = 140; 16.55%)	[46]
South Africa	Cattle	109	57	*B. bigemina* (n = 24; 22.02%) *B. bovis* (n = 33; 30.27%)	[47]
South Africa	Goats	31	0	0.00	[47]
South Africa	Sheep	10	3	*T. ovis* (n = 3; 30.00%)	[47]
South Africa	Cattle	430	278	*B. bigemina* (n = 278; 64.65%), *B. bovis* (n = 151; 35.12%)	[48]
South Africa	Cattle	50	32	*B. bovis* (n = 32; 64.00%)	[49]
South Africa	Cattle	215 755	129	*A. marginale* (n = 129; 60.00%) *Anaplasma* spp. (n = 648; 85.83%)	[50]
South Africa	Cattle	74	39	*A. marginale* (n = 39; 52.70%), *B. bigemina* (n = 2; 2.70%), *Ehrlichia* spp. (n = 14; 18.92%), *T. taurotragi* (n = 26; 35.14%)	[51]
South Africa	Cattle	268	210	*B. bigemina* (n = 204; 76.12%), *B. bovis* (n = 95; 35.45%)	[52]
South Africa	Cattle	250	182	*A. marginale* (n = 182; 72.80%)	[53]
South Africa	Cattle	265	78	*T. parva* (n = 78; 29.43%)	[54]
South Africa	Cattle	70	55	*Anaplasma* spp. (n = 55; 78.57%)	[36]
South Africa	Goats	61	54	*A. ovis* (n = 28; 45.90%), *E. ruminatium* (n = 12; 19.67%), *T. ovis* (n = 14; 22.95%)	[55]
South Africa	Sheep	30	10	*A. ovis* (n = 5; 16.67%), *E. ruminatium* (n = 1; 3.33%), *T. ovis* (n = 4; 13.33%)	[56]

Table 1. *Cont.*

Countries	Hosts	Sample Size	Total No. of Pathogens Detected	Pathogens Detected (No. of Positives, Prevalence (%))	Reference
South Africa	Cattle	1723	48	*E. ruminatium* (n = 48; 2.79%)	[57]
South Africa	Goats	308	17	*E. ruminatium* (n = 17; 5.52%)	[57]
South Africa	Sheep	350	20	*E. ruminatium* (n = 20; 5.71%)	[57]
South Africa	Cattle	81	30	*B. bigemina* (n = 30; 37.04%)	[33]
South Africa	Cattle	170	106	*B. bigemina* (n = 6; 3.53%), *B. bovis* (n = 9; 5.29%), *T. parva* (n = 8; 4.71%), *T. taurotragi* (n = 89; 52.35%)	[57]
South Africa	Cattle	60	50	*B. rossi* (n = 1; 1.67%)*T. mutans* (n = 49; 81.67%), *T. parva* (n = 4; 6.67%), *T. taurotragi* (n = 1; 1.67%), *T. velifera* (n = 42; 70.00%)	[17]
Tanzania	Cattle	354	98	*T. parva* (n = 98; 27.68%)	[58]
Tanzania	Cattle	381	374	*T. parva* (n = 374; 98.16%)	[59]
Tanzania	Cattle	130	124	*T. parva* (n = 124; 95.38%)	[60]
Tanzania	Cattle	960	303	*T. parva* (n = 303; 31.56%)	[61]
Tanzania	Cattle	336	116	*T. parva* (n = 116; 34.52%)	[62]
Tanzania	Cattle	160	39	*T. parva* (n = 39; 24.38%)	[63]
Tanzania	Cattle	245	153	*A. marginale* (n = 39; 15.92%), *B. bigemina* (n = 43; 17.55%), *B. bovis* (n = 11; 4.49%), *T. mutans* (n = 105; 42.86%), *T. ovis* (n = 3; 1.22%), *T. parva* (n = 63; 25.71%), *T. taurotragi* (n = 70; 30.20%)	[64]
Tanzania	Cattle	236	152	*A. marginale* (n = 24; 10.17%), *B. bigemina* (n = 12; 5.08%), *B. bovis* (n = 5; 2.12%), *T. mutans* (n = 90; 38.14%), *T. parva* (n = 81; 34.32%), *T. taurotragi* (n = 73; 30.93%), *T. velifera* (n = 8; 3.39%)	[65]
Tanzania	Cattle	150	105	*T. parva* (n = 105; 70.00%)	[66]
Tanzania	Cattle	64	9	*Theileria* spp. (n = 9; 14.06%)	[67]
Zambia	Cattle	130	21	*B. bigemina* (n = 19; 21.11%) *B. bovis* (n = 2; 2.22%)	[68]
Zambia	Cattle	142	78	*T. parva* (n = 78; 54.93%)	[69]
Zambia	Cattle	472	79	*B. bigemina* (n = 76; 16.10%), *T. parva* (n = 3; 0.64%)	[70]
Zambia	Goats	53	0	0	[70]
Zambia	Cattle	579	181	*Anaplasma* spp. (n = 69; 11.92%), *E. ruminatium* (n = 5; 0.86%), *T. mutans* (n = 94; 16.23%), *T. parva* (n = 4; 0.69%), *T. taurotragi* (n = 4; 0.69%)	[71]
Zambia	Cattle	232	99	*B. bigemina* (n = 24; 10.34%), *T. mutans* (n = 11; 4.74%), *T. parva* (n = 23; 9.91%), *T. taurotragi* (n = 41; 17.67%)	[72]
Zambia	Cattle	299	259	*A. marginale* (n = 77; 25.75%), *B. bigemina* (n = 10; 3.34%), *B. bovis* (n = 23; 7.69%), *T. mutans* (n = 163; 54.52%), *T. parva* (n = 1; 0.33%), *T. velifera* (n = 153; 51.17%)	[73]
Zambia	Cattle	71	34	*A. marginale* (n = 34; 47.89%), *B. bigemina* (n = 16; 22.54%), *T. parva* (n = 16; 22.54%)	[74]
Zimbabwe	Cattle	94	33	*B. bigemina* (n = 33; 35.11%), *B. bovis* (n = 27; 28.72%)	[75]

Table 2. Sub-group analysis for infection rates of tick-borne pathogens associated with animal hosts, pathogen genera, diagnostic technique, study years and countries.

Risk Factors	Number of Studies	Pooled Prevalence Estimates			Measure of Heterogeneity		Q-*p*	Publication Bias
		Sample Size	No. of Positive	Prevalence 95% CI	Q	I²		Begg and Mazumdar Rank *p*-Value
Overall Animals	48	12693	5172	52.2 (43.9–60.3)	2820.792	98.33	0.609	0.065
Animal hosts								
Cattle	45	12693	5172	51.2 (42.9–59.4)	2491.04	98.23	0.779	0.056
Goats	8	663	236	29.9 (7.3–69.9)	252.68	97.23	0.325	0.310
Sheep	5	483	109	45.4 (9.4–87.0)	146.22	97.26	0.861	0.312

Table 2. *Cont.*

Risk Factors	Number of Studies	Pooled Prevalence Estimates			Measure of Heterogeneity		Q-p	Publication Bias
		Sample Size	No. of Positive	Prevalence 95% CI	Q	I²		Begg and Mazumdar Rank *p*-Value
Genus *Anaplasma*								
A. bovis	2	565	5	0.88	-	-	-	-
A. centrale	4	1148	146	14.7 (5.9–32.0)	69.01	95.65	0.001	0.500
A. marginale	14	2982	1264	45.9 (31.3–61.3)	618.20	97.90	0.605	0.351
A. phagocytophilum	2	317	8	2.52	-	-	-	-
Anaplasma spp.	7	2216	1126	45.6 (17.9–76.3)	760.30	99.21	0.797	0.440
Genus *Babesia*								
B. bigemina	22	4393	1280	20.8 (12.4–32.6)	1007.80	97.92	0.000	0.068
B. bovis	14	2733	723	20.3 (12.7–30.9)	373.29	96.52	0.000	0.070
Genus *Ehrlichia*								
E. ruminantium	5	2936	118	4.2 (1.6–10.2)	74.03	94.60	0.000	0.500
Ehrlichia spp.	2	551	43	7.80	-	-	-	-
Genus *Theileria*								
Theileria spp.	1	64	9	14.06	-	-	-	-
T. mutans	10	2591	831	29.1 (17.5–44.4)	369.35	97.56	0.009	0.210
T. parva	20	6288	1712	25.0 (17.6–34.1)	687.51	97.24	0.000	0.097
T. velifera	6	1236	549	43.0 (26.4–61.4)	135.20	96.30	0.459	0.286
Diagnostic technique								
nPCR	14	3815	2006	61.5 (45.6–75.2)	799.92	98.38	0.155	0.104
PCR	28	5432	2291	43.6 (34.8–52.8)	863.936	96.88	0.172	0.376
qPCR	4	2534	475	31.0 (6.7–73.8)	537.17	99.44	0.393	0.248
RLB	7	1428	863	63.0 (42.0–80.0)	201.38	97.02	0.222	0.440
RT-PCR	2	1046	194	18.55	-	-	-	-
htPCR	1	117	86	73.50	-	-	-	-
Study year								
1990–2000	1	276	2	0.72	-	-	-	-
2001–2010	9	2023	1170	63.6 (49.1–75.9)	273.97	97.08	0.066	0.267
2011–2020	21	5085	2586	57.3 (46.4–67.6)	844.80	97.63	0.187	0.040
Study countries								
Angola	4	338	178	54.3 (21.9–83.5)	85.86	96.51	0.814	0.248
Botswana	2	705	137	19.43	-	-	-	-
Mozambique	5	1072	677	62.9 (25.3–89.5)	255.31	98.43	0.521	0.312
South Africa	18	5543	1922	52.2 (37.6–66.4)	1212.40	98.60	0.772	0.367
Tanzania	10	3016	1474	57.8 (42.2–72.0)	432.85	97.92	0.326	0.020
Zambia	7	1925	751	41.7 (24.1–61.7)	330.51	98.18	0.417	0.226
Zimbabwe	1	94	33	35.11	-	-	-	-

htPCR: High throughput qPCR; PCR: conventional polymerase chain reaction; nPCR: nested PCR; qPCR: real time polymerase chain reaction; RT-PCR: reverse transcription-polymerase chain reaction; RLB: reverse line blot hybridization.

2.3. Pooling, Heterogeneity, and Sub-Group Analysis

2.3.1. Prevalence in Animals Based on Host, Study Years, Countries, Diagnostic Technique and Species of Tick-Borne Pathogens

The overall pooled prevalence estimate (PPE) of tick-borne pathogens (TBPs) in animals was 52.2% [(95% CI: 43.9–60.3%); Q = 2820.792; I² = 98.33; *Q-p* = 0.609] (Table 2). The subgroup analysis by animal hosts revealed that the highest PPE in cattle was 51.2% [(95% CI: 42.9–59.4%); Q = 2491.04; I² = 98.23; *Q-p* = 0.779], followed by sheep [45.4% (95% CI: 9.4–87.0%); Q = 146.22; I² = 97.26; *Q-p* = 0.861], and goats had the lowest PPE [29.9% (95% CI: 7.3–69.9%); Q = 252.68; I2 = 97.23; *Q-p* = 0.325] (Table 2). Only one study was

eligible for inclusion within the study period spanning from 1990 to 2000 with a PPE of 0.72%, while 21 studies were included within the period from 2011 to 2020 with a PPE of 57.3% (Table 2). Lastly, a PPE of 63.6% was observed in the period from 2001 to 2010. Seven studies used RLB diagnostic methods for the detection of tick-borne pathogens and these diagnostic methods recorded the highest pooled rate as 63.0% (95% CI: 42.0–80.0), followed by nPCR with 61.5 (95% CI: 45.6–75.2). PCR was recorded with a pooled prevalence of 43.6% (95% CI: 34.8–52.8) from twenty-eight studies. Statistics are recorded in Table 2. The forest plots that show the point estimates for the individual studies that describe the occurrence of *Anaplasma marginale*, *Babesia bigemina*, *B. bovis*, and *Theileria parva* are presented (Supplementary Figures S1–S4).

2.3.2. Prevalence of Tick-Borne Pathogens in Different Species of Ticks

Different TBPs were detected and reported in tick species collected from animals within the SADC region at varying prevalence (Table 3). The overall PPE of TBPs in tick populations was 10.7% [(95% CI: 5.7–19.1%); $Q = 2132.53$; $I^2 = 99.16$; Q-$p = 0.086$] (Table 4). The subgroup analysis of bacterial TBPs is shown in Table 4.

Table 3. Characteristics of all eligible studies reporting the occurrence of tick-borne pathogens in ticks collected from domestic ruminants across the Southern African Developing Community region.

Countries	Hosts	Tick Species	Molecular Technique	Sample Size	Counts of Detected Pathogens in Ticks	Pathogens Detected (No. of Positives, Prevalence (%)	Reference
Angola	Cattle	*A. variegatum, R. decoloratus*	PCR	116	6	*R. africae* (n = 5; 4.31%), *T. mutans* (n = 1; 0.86%)	[30]
Angola	Cattle, goats, sheep	*R. compositus*	PCR, RLB	2963	43	*E. ruminatium* (n = 43; 1.45%)	[32]
Comoros	Cattle, Goats	*A. variegatum, R. appendiculatus, R.(B). microplus*	PCR	512	94	*R. africae* (n = 94; 18.36%)	[76]
Madagascar	Cattle, Goats	*H. simplex, R. microplus*	PCR	235	60	*R. africae* (n = 60; 26.67%)	[77]
Madagascar	Cattle	*A. variegatum, R. microplus*	PCR	499	312	*A. marginale* (n = 311; 62.32%), *A. ovis* (n = 1; 0.15%)	[78]
Mozambique	Cattle	*A. variegatum, R. microplus*	PCR	646	5	*R. africae* (n = 4; 0.62%) *T. velifera* (n = 1; 0.15%)	[79]
South Africa	Cattle, goats, sheep	*R. appendiculatus, R. decoloratus, R. e. evertsi*	PCR	1200	26	*E. ruminatium* (n = 19; 1.58%), *A. bovis* (n = 1; 0.25%), *A. marginale* (n = 2; 0.15%), *A. ovis* (n = 3; 0.33%), *B. caballi* (n = 1; 0.25%)	[80]
South Africa	Cattle, sheep	*A. hebraeum, H.m. rufipes, R. appendiculatus, R. (B.) decoloratus, R. e. evertsi*	PCR	7364	58	*B. bigemina* (n = 4; 0.31%), *Babesia* spp. (n = 1; 0.38%), *E. ruminatium* (n = 5; 2.15%), *E. ovina* (n = 2; 0.17%), *Ehrlichia* spp. (n = 8; 0.61%), *T. bicornis* (n = 7; 0.75%), *T. buffeli* (n = 7; 0.45%), *T. mutans* (n = 2; 0.18%), *T. ovis* (n = 2; 0.22%), *T. separata* (n = 4; 0.44%), *T. taurotragi* (n = 3; 0.32%), *Theileria* spp. (n = 13; 0.71%)	[81]
South Africa	Cattle, sheep	*A. hebraeum, R. appendiculatus, R. decoloratus, R. e. evertsi*	PCR	130	24	*A. marginale* (n = 5; 3.85%), *E. ruminatium* (n = 2; 1.54%), *Rickettsia* spp. (n = 10; 7.69%), *T. mutans* (n = 4; 3.08%), *T. taurotragi* (n = 3; 2.31%)	[82]
South Africa	Cattle, goats, sheep	*A. hebraeum, R. appendiculatus, R. decoloratus, R. e. evertsi, R. sanguineus*	PCR	760	16	*Ehrlichia* spp. (n = 16; 2.10%)	[83]

Table 3. *Cont.*

Countries	Hosts	Tick Species	Molecular Technique	Sample Size	Counts of Detected Pathogens in Ticks	Pathogens Detected (No. of Positives, Prevalence (%))	Reference
South Africa	Cattle	*R. sanguineus*	PCR	100	10	*A. phagocytophilum* (n = 10; 10%)	[85]
South Africa	Cattle, goats, sheep	*A. hebraeum, H. truncatum, R. appendiculatus, R. e. evertsi, R. microplus, R. simus*	PCR	903	60	*Rickettsia* spp. (n = 60; 6.64%)	[84]
South Africa	Cattle, goats, sheep	*A. hebraeum*	PCR	1403	344	*E. ruminatium* (n = 344; 24.52%)	[57]
South Africa	Goats	*A. hebraeum*	PCR	630	47	*E. ruminatium* (n = 19; 3.02%) *R. africae* (n = 28; 4.44%)	[86]
Tanzania	Cattle, Goats	-	PCR	819	0	-	[87]
Tanzania	Cattle	*A. gemma, R. appendiculatus, R. praetextatus, R. pulchellus*	PCR	527	28	*A. marginale* (n = 28; 5.31%)	[88]
Tanzania	Cattle	*A. gemma, A. lepidum, A. marmoreum, A. variegatum, H. impeltatum, R. pulchellus*	PCR	263	160	*Babesia* spp. (n = 7; 2.66%), *Ehrlichia* spp. (n = 6; 2.28%), *Rickettsia* spp. (n = 133; 50.57%), *Theileria* spp. (n = 14; 5.32%)	[89]
Zambia	Cattle	*A. variegatum*	RLB	5288	1	*E. ruminatium* (n = 1; 0.02%)	[73]
Zambia	Cattle	*R. appendiculatus*	PCR	74	10	*T. parva* (n = 10; 13.51%)	[74]
Zimbabwe	Cattle	*H. truncatum, R. e. evertsi*	PCR	1141	288	*E. ruminatium* (n = 288; 25.24%)	[90]
Zimbabwe	Cattle	*R. appendiculatus*	PCR	36	18	*B. bigemina* (n = 12; 33.33%), *B. bovis* (n = 6; 16.67%)	[75]

Table 4. Sub-group analysis for infection rates of tick-borne pathogens detected in ticks collected from different domestic ruminants.

Risk Factors	Number of Studies	Pooled Prevalence Estimates			Measure of Heterogeneity		Q-p	Publication Bias
		Sample Size	Number of Positive	Prevalence 95% CI (%)	Q	I^2		Begg and Mazumdar Rank *p*-Value
Overall ticks	20	18355	1601	7.7 (4.0–14.3)	2310.69	99.18	0.000	0.060
Genus *Anaplasma* *A. marginale*	4	2428	348	6.8 (0.6–45.2)	333.05	99.10	0.034	0.248
Genus *Ehrlichia* *E. ruminantium* *Ehrlichia* spp.	8 3	3719 1543	701 31	4.6 (2.2–9.1) 2.1 (1.4–3.3)	347.46 3.02	97.98 33.72	0.000 0.000	0.161 0.301
Genus *Rickettsia* *R. africae* *Rickettsia* spp.	5 3	978 859	185 203	18.0 (7.4–37.5) 39.0 (4.0–90.8)	104.23 136.03	96.16 98.53	0.003 0.749	0.164 0.059
Genus *Theileria* *T. mutans*	3	1193	7	2.6 (0.2–31.2)	23.58	91.52	0.012	0.301

2.3.3. Prevalence of Tick-Borne Pathogens in Different Species of Ticks

Thirty tick species belonging to 8 genera, namely, *Rhipicephalus* (n = 10 species), *Amblyomma* (n = 9 species), *Heamaphysalis* (n = 3 species), *Boophilus* (n = 2 species), *Hyalomma* (n = 2 species), *Ixodes* (n = 2 species), *Margaropus* (n = 1 species), and *Otobius* (n = 1 species) were reported in the studies from different SADC countries (Table 5). The PPE of TBPs in 22 tick species from five different genera revealed pathogens to be more prevalent in the tick genus *Amblyomma* at 25.0% [(95% CI: 14.7–39.1%); Q = 598.25; I^2 = 97.66; Q-p = 0.001], as

compared to the genus *Rhipicephalus* at 11.7% [(95% CI: 4.7–26.2%); Q = 786.55; I^2 = 98.47; Q-p = 0.000]. Other tick genera, including *Boophilus*, *Hyalomma*, and *Haemaphysalis*, expressed a PPE of 50%, 13.1%, and 10.5% for TBPs, respectively (Table 5).

Table 5. Pooled prevalence estimates and risk factor associated with ticks species and tick-borne pathogen infections in animal ticks.

| Risk Factors | Number of Studies | Pooled Prevalence Estimates | | | Measure of Heterogeneity | | Q-p | Publication Bias |
		Sample Size	No. of Positive	Prevalence 95% CI (%)	Q	I^2		Begg and Mazumdar Rank *p*-Value
Genus *Amblyomma*	15	3987	959	25.0 (14.7–39.1)	598.25	97.66	0.001	0.200
A. chabaudi	1	2	2	100.00	-	-	-	-
A. gemma	2	79	20	25.32				
A. hebraeum	7	2344	456	14.2 (8.9–21.8)	64.23	90.66	0.000	0.440
A. lepidum	1	42	8	19.05	-	-	-	-
A. marmoreum	1	11	2	18.18	-	-	-	-
A. pomposum	1	617	43	6.97	-	-	-	-
A. variegatum	7	3713	431	43.9 (10.1–84.4)	250.42	97.60	0.804	0.440
Genus *Haemaphysalis*	1	19	2	10.53	-	-	-	-
H. simplex	1	19	2	10.53	-	-	-	-
Genus *Hyalomma*	2	909	119	13.1	-	-	-	-
H.m. rufipes	2	582	89	15.29	-	-	-	-
H. truncatum	1	327	20	6.12	-	-	-	-
Genus *Rhipicephalus*	14	8730	522	8.0 (3.2–18.6)	841.80	98.46	0.000	0.162
R. appendiculatus	9	899	40	3.7 (1.6–8.3)	46.25	82.70	0.000	0.266
R. (B.) decoloratus	3	424	36	36.9 (3.1–91.5)	63.91	96.87	0.000	0.30
R. compositus	2	181	7	3.87	-	-	-	-
R. decoloratus	2	42	12	28.57	-	-	-	-
R.(B). microplus	2	312	14	4.49	-	-	-	-
R. e. evertsi	5	1718	234	7.4 (1.1–35.8)	317.76	98.74	0.011	0.312
R. microplus	3	693	174	15.4 (1.1–75.5)	173.07	98.84	0.238	0.301
R. praetextatus	1	23	2	8.70	-	-	-	-
R. pulchellus	1	22	6	27.27	-	-	-	-
R. sanguineus	2	280	15	5.36	-	-	-	-

The analysis of tick species harbouring TBPs showed that *A. variegatum* was the species that harboured the most pathogen infections at 43.9% [(95% CI: 10.1–84.4%); Q = 250.42; I^2 = 97.60; Q-p = 0.804]; followed by *R. microplus* at 15.4% [(95% CI: 1.1–75.5); Q = 173.07; I^2 = 98.84; Q-p = 0.238]; then, *A. hebraeum* at 14.2% [(95% CI: 8.9–21.8%); Q = 64.23; I^2 = 90.66; Q-p = 0.000]; *R. e. evertsi* at 7.4% [(95% CI: 1.1–35.8%); Q = 317.76; I^2 = 98.74; Q-p = 0.011]; and *R. appendiculatus* at 5.5% [(95% CI: 2.8–10.4%); Q = 25.93; I^2 = 73.00; Q-p = 0.000]. The percentages of tick species infected with TBPs, in descending order, was as follows: the *A. chabaudi* (100.0%), followed by *B. decoloratus* (60.0%), *B. microplus* (37.5%), *R. decoloratus* (28.6%), *R. pulchellus* (27.3%), *A. gemma* (25.3%), *A. lepidum* (19.1%), *A. marmoreum* (18.2%), *H. m. rufipes* (15.3%), *H. simplex* (10.5%), *R. praetextatus* (8.7%), *A. pomposum* (7.0%), *H. truncatum* (6.1%), *R. (B.) decoloratus* (5.9%), *R. sanguineus* (5.4%), *R. compositus* (3.9%), and *R.(B). microplus* (2.7%) [Table 6]. South Africa appeared to be the country with highest TBP prevalence in ticks, as compared to other countries in the SADC region, since most of their tick studies tested negative for tick-borne pathogens (Table 4, Figure 2).

Table 6. Search strategies.

S/No.	Source	Query/Search String	Results
1	PubMed	Ticks and tick-borne pathogens in Southern Africa; Prevalence of "Anaplasma" "Babesia" "Ehrlichia" and/or "Theileria"	56
2	Science direct	Ticks and tick-borne pathogens in Southern Africa; Prevalence of "Anaplasma" "Babesia" "Ehrlichia" and/or "Theileria"	751
3	Google scholar	Ticks and tick-borne pathogens in Southern Africa; Prevalence of "Anaplasma" "Babesia" "Ehrlichia" and/or "Theileria"	31,700
4	AJOL	Ticks and tick-borne pathogens in Southern Africa; Prevalence of "Anaplasma" "Babesia" "Ehrlichia" and/or "Theileria"	244
5	Springer Link	Ticks and tick-borne pathogens in Southern Africa; Prevalence of "Anaplasma" "Babesia" "Ehrlichia" and/or "Theileria"	743

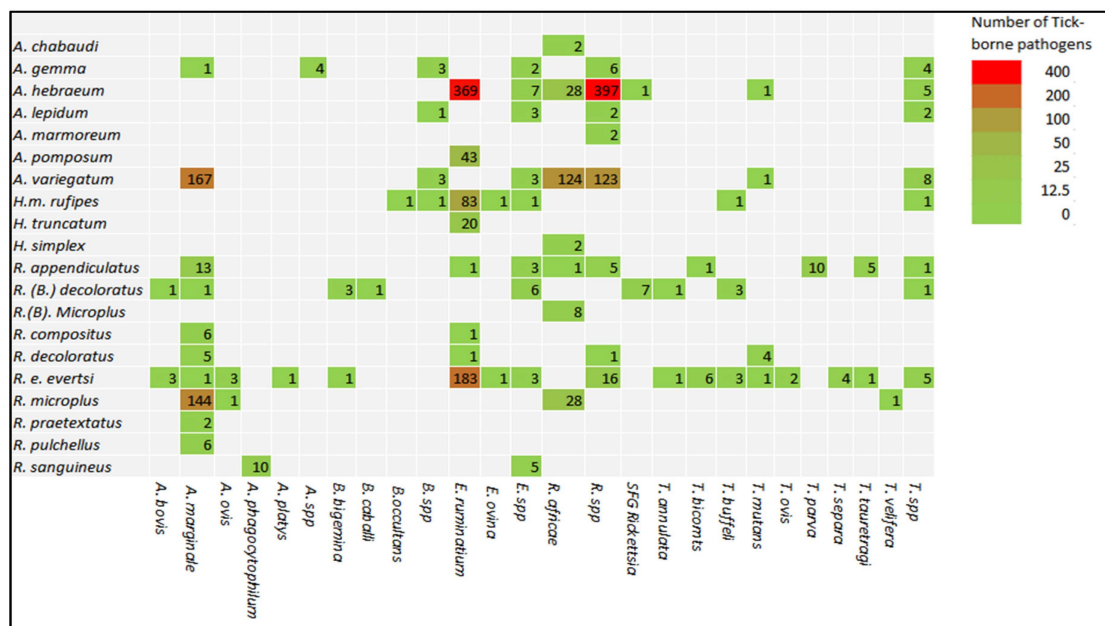S/No. = Searching number.

Figure 2. Heat map for linked relationship between ticks and tick-borne pathogens in the Southern African Developing Community region.

2.3.4. Assessment for Publication Bias in Studies Involving Domestic Ruminant Animals

A funnel plot of standard error by logit event rate together with the Begg and Mazumdar rank correlation test were used to assess publication bias. Our data analyses showed no evidence of publication bias for almost all risk factors for animal studies, except for study period 2010–2021 (Table 2, Figure 3) and the country Tanzania, where significant bias was observed in regard to asymmetries of the funnel plots and *p*-values of 0.040 and 0.020, respectively (Table 2, Figure 4).
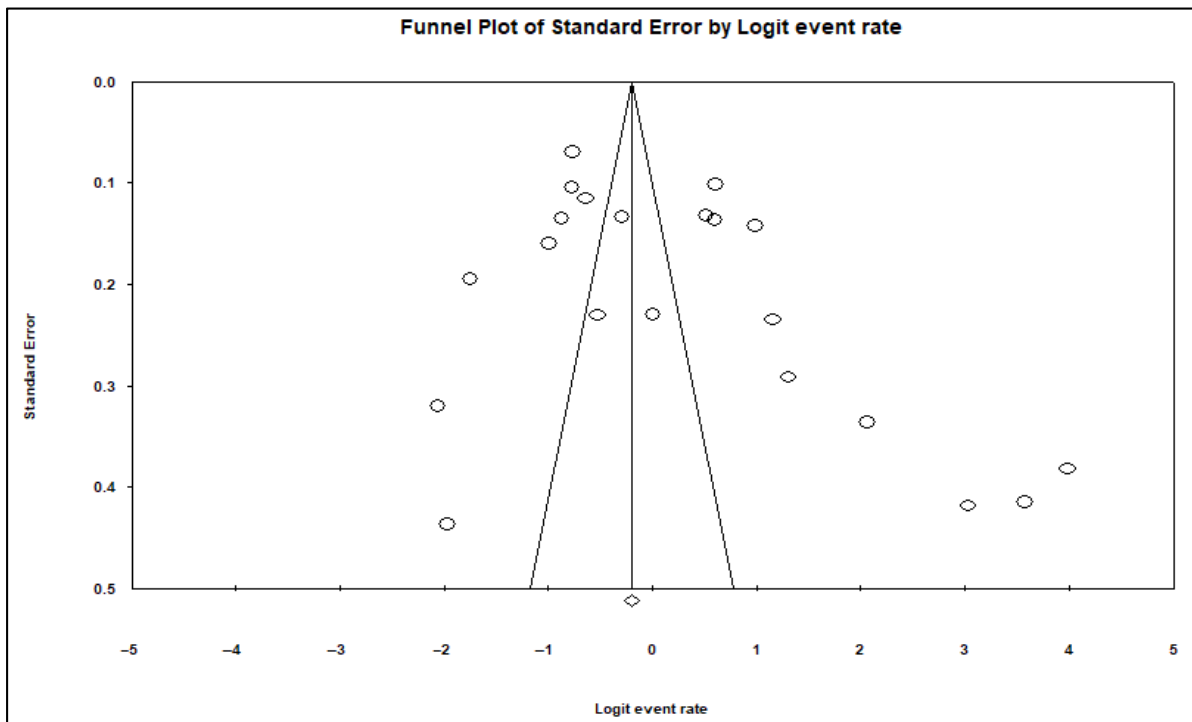
Figure 3. A funnel plot of subgroup studies tested reported positive detection of tick-borne pathogens in livestock for 2011–2021 year interval/period.
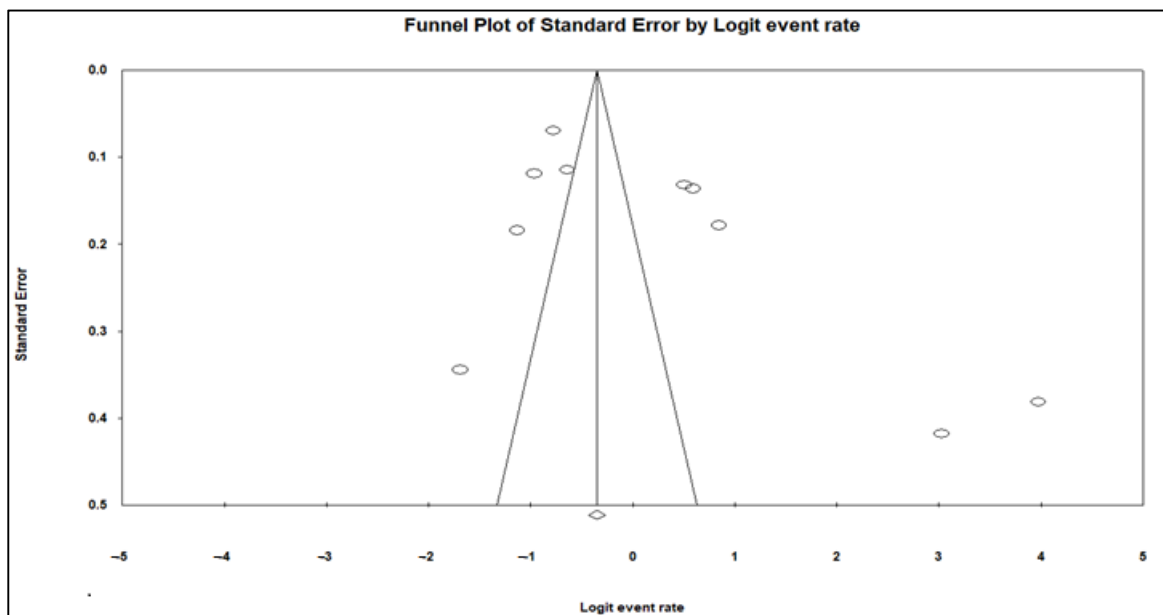

Figure 4. A funnel plot subgroup studies that positively detected tick-borne pathogens from livestock in Tanzania.

3. Discussion

3.1. Ticks

Tick identification and prevalence are significant factors in estimating the abundance of tick species in a population and in quantifying the prevalence of TBPs of public and animal health concern [91,92]. Tick-borne pathogens in African domestic ruminants are

complex, with several tick species feeding on various animals to facilitate the transmission of numerous microorganisms [93].

In this study, a total of 26 tick species belonging to five genera of *Ixodidae*, namely, *Amblyomma*, *Hyalomma*, *Haemaphysalis*, and *Rhipicephalus* (including *Boophilus*), were recorded, with the *Rhipicephalus* genus being the most abundant. Similar findings were recorded in the Caribbean, where the genus *Rhipicephalus* was more prevalent as compared to other tick genera [94,95].

We also observed that *Amblyomma* ticks harboured more TBPs, as compared to the genus *Rhipicephalus*. These results are slightly similar to those reported in Kenya [96] and in Guinea and Liberia [97]. Specifically, *A. variegatum* was the most infected tick vector, while *R. appendiculatus* was the least; this finding is consistent with a previous report from Ethiopia [98]. In contrast, the authors of [99] reported a lower prevalence of tick-borne pathogens in *A. variegatum* in Oromia, Ethiopia. The high prevalence of tick-borne pathogens on *A. variegatum* ticks might suggest a public health concern to people living in the areas of sample collection if bitten by infected ticks, since this tick is known to transmit pathogens to both animals and humans [97].

There is a significant public and animal health risk associated with tick host preference, as they harbour zoonotic pathogens, including *Anaplasma* spp., *Babesia* spp., *Ehrlichia* spp., and *Rickettsia*, which can be transmitted at the interface between domestic animals, wildlife, and humans [100]. The overall PPE of TBPs in different ticks was below 20%, similar to previous reports in Turkey [101], Japan [102], and Greece [103]. However, higher prevalences of TBPs harboured by ticks was reported in Sudan [42.7%] [104] and Pakistan [35.1%] [105]. Spotted fever group is a causative agent of the rickettsioses belonging to the family *Rickettsiaceae* [106]. Ticks in the genus *Amblyomma* are known to be reservoirs and transmitters of *Rickettsia* spp. in cattle and humans in Africa [107]. According to Althaus et al. [108], *Rickettsia africae* is reportedly the most common species of rickettsial pathogens detected in ticks and humans in some parts of the African continent and is responsible for ~11% of African tick bite fever cases in tourists from South Africa.

3.2. Tick-Borne Pathogens in Different Animal Host

According to the analysed, published articles in the current study, TBPs have been detected from domestic animals in Angola, Botswana, Mozambique, South Africa, Tanzania, Zambia, and Zimbabwe with an overall PPE of 52.2%, which is a representative portion of the SADC region. This PPE is higher than that reported from Algeria (Central Africa), with prevalence of less than 20% [109], but was slightly lower than the 62.9% reported in Uganda (Eastern Africa) [110]. This high prevalence in SADC countries might be due to farm management, micro-climate patterns, tick distribution, and livestock breeds [111,112].

The PPE of TBPs in domestic ruminant as observed in this study was similar to that reported from the Caribbean, where cattle had a higher prevalence compared to sheep and goats [113]. In contrast, Bell-Sakyi et al. [114] reported TBPs from cattle to have a lesser prevalence compared to sheep and goats in Ghana. This variation in host prevalence might be associated with the livestock husbandry system practiced in most SADC countries, where cattle farming predominates that of small ruminants [115].

The genus *Anaplasma* with causal agents of anaplasmosis in cattle had a higher prevalence (45.6%) for the SADC countries in this study, as compared to the 5.3% and 22.6% that were previously reported from Uganda and Senegal, respectively [87,116]. Evidence of a similar prevalence to the current study was reported in Italy [50%] and Iran [77%] [117,118]. However, results according to species level revealed that *A. marginale* was the most prevalent species of *Anaplasma*, which is similar to reports from Turkey [119] and Iran [120]. In contrast, Mohammadian et al. [121] and Salehi-Guilandeh et al. [122] reported *A marginale* to be a less prevalent species in cattle, compared to other *Anaplasma* species in the west and northern regions of Iran.

Babesia are known to be pathogenic to cattle and can also pose a serious public health risk to humans [123]. The prevalence of *B. bigemina* was slightly higher as compared to

B. bovis. Similar findings were observed in Brazil (34% for *B. bigemina* and 20.4% for *B. bovis*) [124] and Colombia (24.2% for *B. bigemina* and 14.4% for *B. bovis*) [125]. These findings can be explained by the observations of Kocan [126], who reported that higher temperatures hinder or terminate the synthesis of the *B. bigemina* pathogen in cattle ticks.

In the current study, the PPE of 4.2% for *E. ruminantium* is relatively similar to the 4.5% and 6.6% prevalences that were reported in Western Uganda [116] and Cameroon, respectively [127]. The PPE of the current study is, however, higher than the 0.5% and 1.1% prevalences reported in South-western Ethiopia and Nigeria, respectively [128,129]. The low prevalence might be due to colostrum and indigenous breed genes that can influence some level of resistance to *Ehrlichia* pathogens in domestic animals [130,131].

The species of the genus *Theileria* recorded from the studies used in this systematic review and meta-analysis includes *T. velifera* (43%) as the most prevalent, followed by *T. mutans* (29.1%), and *T. parva* (25.0%). In contrast, *T. velifera* was reported to be less prevalent than *T. parva* in Kenya and Uganda, with a prevalence of *T. velifera* at 1.3% and *T. parva* at 1.9% in Kenya, and *T. velifera* at 11.8% and *T. parva* 69.4% in Uganda. [132,133]. However, other studies reported *T. velifera* to be less prevalent than *T. mutans* in Southern Sudan [*T. velifera* 45.3%, *T. mutans* 73%] [134] and in Ethiopia [*T. velifera* 4%, *T. mutans* 8%] [135]. Our findings are in congruence with the results reported by Byaruhanga et al. [136], where the *T. velifera* [71.3%] prevalence was higher than *T. parva* [2.9%] in the Karamoja region, Uganda, and *T. velifera* [40%] was higher compared to *T. mutans* [25.7%] in Lambwe Valley, Kenya [137].

The findings on different molecular techniques revealed that the RLB technique was the most sensitive of all molecular-based methods. This is due to the fact that RLB assay is able to detect various TBPs from the same specimen simultaneously, unlike conventional PCR [138,139]. Similar results in accordance with current data were reported in a study detecting TBPs in Western Kenya using RLB and qPCR techniques, in which RLB was more sensitive compared to qPCR [93]. Furthermore, a study on small ruminant blood samples in Turkey reported that RT-PCR is more sensitive than the RLB method at detecting TBPs [24].

With regard to changes in prevalence over time, a declining trend in tick-borne pathogens prevalence was observed over the course of the 10-year intervals in our study, from the period 2001–2010 to the period 2011–2020 among cattle in SADC regions. The decline over time witnessed in this study suggests that there might have been some improvements by countries' agricultural sectors (government and farmers) in tick, TBPs, and TBPDs control measures, such as proper use of acaricides during spraying and dipping.

3.3. Limitations

This regional approach provides statistical evidence and a clearer understanding of the spatial distribution of ticks and TBPs in the SADC region, which will assist in identifying countries in which there is a lack of scientific evidence. Future research may prioritize areas of emphasis for better understanding and consolidation of data to develop prevention and control strategies. We also highlighted the link between ticks and their pathogens by observing the interesting roles of this link from an epidemiological point of view. This systematic review and meta-analysis have multiple limitations, such as: (i) there was a small number of domestic ruminant studies conducted in the SADC; therefore, a subgroup analysis was impossible to conduct due to few eligible articles. This lack of small domestic ruminants data may be due to the influences of study location, farming activities of countries, belief or myths, sample sizes, availability of animals, and study designs of the individual studies. (ii) Most studies did not consider demographic characteristics, such as life stage of ticks, age of animals, or sex of animals and ticks. (iii) There was a large gap of study outputs between the study period from one publication from 1990–2000 and 21 publications in 2010–2020. (iv) There is also lack of published studies for TBPs using molecular diagnostic techniques from many countries, such as the Democratic Republic of Congo, Lesotho, Mauritius, Namibia, Seychelles, and Swaziland, and as a result, the analysis is not entirely representative of the entire SADC region. In Comoros and Madagascar, there were no

articles published in which blood samples were screened for TBPs, while Botswana and Malawi did not have tick sample studies.

4. Materials and Methods

4.1. Search Strategy and Criteria

Literature searches were conducted on PubMed, Science Direct, and Google Scholar for articles published in the English language from 1 January 1980 until 22 March 2021, with content containing information on the prevalence or epidemiology of tick-borne pathogens across SADC countries in domestic animals. The search keywords were Ticks and Tick-borne pathogens in Southern Africa; Prevalence of "*Anaplasma*" "*Babesia*" "*Ehrlichia*" and/or "*Theileria*". Keywords were used individually or in combination with the "AND" and/or "OR" operators (Table 6). None of the authors of the original studies were contacted for additional information and no attempt was made to retrieve unpublished articles. Titles and abstracts were scanned and relevant full text articles were downloaded and obtained through library resources and online platforms.

4.2. Inclusion and Exclusion Criteria

Articles were included only if they fulfilled the following inclusion criteria: cross sectional (prevalence) study conducted within the SADC region; involved invertebrate (ticks) and vertebrate host (cattle, goats and sheep); involved ticks and/or blood sample collection; exact total numbers and positive cases were clearly provided; involved the use of a molecular-based technique; sample size (>25 for enabling statistical calculations); written in English. Studies without the above-mentioned characters, such as reviews, experimental studies, non-domestic ruminant studies, insufficient data analyses, studies with lower sample sizes, and studies not written in English were all excluded.

4.3. Data Extraction

The data extraction protocol was prepared and evaluated by all authors. The data extraction protocol consists of the names of the authors and countries, hosts, total sample sizes, number of positive cases, estimated prevalence, species of blood pathogens, tick species, and molecular diagnostic technique. Moreover, studies that were conducted in more than one country and those that had both animal and tick studies simultaneously were separated accordingly.

Titles and abstracts derived through primary electronic search were thoroughly assessed for possibility of inclusion, based on the study type (prevalence of ticks and tick-borne pathogens of domestic ruminants) in the SADC region. From each eligible study, the following data were extracted, based on the performed software (Excel, Microsoft, 2016) format: author, study area, host, method used, study year, sample size, positive samples, different tick-borne pathogen species, and different tick species. All data were extracted using a standardized extraction form. For duplicate studies, only one study was selected. The extracted data were cross checked with the included papers, then modifications and editions of mistyped data were made when necessary.

4.4. Meta-Analytic Procedures

The current meta-analysis was conducted using Comprehensive Meta-Analysis software (CMA) version 3.0 software [140]. The pooled prevalence estimates and 95% CIs were calculated using random-effects models. Statistical heterogeneity between studies was measured by I^2 statistic; $I^2 > 50\%$ was defined as high heterogeneity [141]. Publication bias was measured using funnel plots to test the symmetry and the Begg's and Mazumdar rank correlation test [142].

5. Conclusions

The highest PPE of TBPs in domestic animals in the SADC is recorded in Mozambique, which has warm subtropical and tropical climates, while the country of Botswana had the

lowest PPE and a semi-arid climate. The major TBPs with high PPE in the SADC region for bacteria and piroplasmids include *A. marginale* and *T. velifera*. The RBL is more sensitive in detecting TBPs from blood samples as compared with other molecular techniques. The most prevalent TBP detected from ticks was *R. africae*, whilst *Rhipicephalus* ticks was the most prevalent in livestock. A higher prevalence rate of TBPs in the SADC was observed in domestic animal blood compared to tick species. Some of the TBDs are transboundary and require cooperation between neighbouring countries for their effective control. There is, therefore, a requirement for consolidated research into regional ticks and TBDs between SADC countries that would enable a united effort in documenting the prevalence and understanding of ticks and TBDs in the region.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/pathogens11080929/s1, Figure S1: Forest plot showing the pooled estimates of *Anaplasma marginale* in South Africa. The squares demonstrate the individual point estimate. The diamond at the base indicates the pooled estimates from the overall studies. Figure S2: Forest plot showing the prevalence of *Babesia bigemina* in South Africa. The squares demonstrate the individual point estimate. The diamond at the base indicates the pooled estimates from the overall studies. Figure S3: Forest plot showing the pooled estimates of *Babesia bovis* in South Africa. The squares demonstrate the individual point estimate. The diamond at the base indicates the pooled estimates from the overall studies. Forest plot showing the prevalence of *Theileria parva* in South Africa. Figure S4: The squares demonstrate the individual point estimate. The diamond at the base indicates the pooled estimates from the overall studies.

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References

1. Adenubi, O.T.; Ahmed, A.S.; Fasina, F.O.; McGaw, L.J.; Eloff, J.N.; Naidoo, V. Pesticidal plants as a possible alternative to synthetic acaricides in tick control: A systematic review and meta-analysis. *Ind. Crop. Prod.* **2018**, *123*, 779–806. [CrossRef]
2. Wu, X.B.; Na, R.H.; Wei, S.S.; Zhu, J.S.; Peng, H.J. Distribution of tick-borne diseases in China. *Parasites Vectors* **2013**, *6*, 119. [CrossRef] [PubMed]
3. Gray, J.S. *Ixodes ricinus* seasonal activity: Implications of global warming indicated by revisiting tick and weather data. *Int. J. Med. Microbiol.* **2008**, *298*, 19–24. [CrossRef]
4. Sirotkin, M.B.; Korenberg, E. Influence of abiotic factors on different developmental stages of the taiga (*Ixodes persulcatus*) and European forest (*Ixodes ricinus*) ticks. *Zool. Zhurnal* **2018**, *97*, 379–396. [CrossRef]
5. Uspensky, I. Infections with Natural Focality Transmitted by *Ixodid* Ticks. *Am. Entomol.* **2016**, *62*, 260–262. [CrossRef]
6. Vorou, R.; Pierroutsakos, I.N.; Maltezou, H.C. Crimean-Congo hemorrhagic fever. *Curr. Opin. Infect. Dis.* **2007**, *20*, 495–500. [CrossRef]
7. Apanaskevich, D.A.; Horak, I.G.; Matthee, C.; Matthee, S. A new species of *Ixodes* (*Acari*: *Ixodidae*) from South African mammals. *J. Parasitol.* **2011**, *97*, 389–398. [CrossRef]
8. Dantas-Torres, F.; Chomel, B.B.; Otranto, D. Ticks and tick-borne diseases: A One Health perspective. *Trends Parasitol.* **2012**, *28*, 437–446. [CrossRef]
9. Fernandes, É.K.; Bittencourt, V.R.; Roberts, D.W. Perspectives on the potential of entomopathogenic fungi in biological control of ticks. *Exp. Parasitol.* **2012**, *130*, 300–305. [CrossRef]

10. Horak, I.G.; Camicas, J.L.; Keirans, J.E. The *Argasidae, Ixodidae* and *Nuttalliellidae* (*Acari*: *Ixodida*): A world list of valid tick names. *Exp. Appl. Acarol.* **2003**, *28*, 27–54. [CrossRef]

11. Horak, I.G.; Nyangiwe, N.; De Matos, C.; Neves, L. Species composition and geographic distribution of ticks infesting cattle, goats and dogs in a temperate and a subtropical coastal region of south-eastern Africa. *Onderstepoort J. Vet. Res.* **2009**, *76*, 263–278. [CrossRef]

12. Guglielmone, A.A.; Robbins, R.G.; Apanaskevich, D.A.; Petney, T.N.; Estrada-Pena, A.; Horak, I.G.; Shao, R.; Barker, S.C. *The Argasidae, Ixodidae and Nuttalliellidae (Acari: Ixodida) of the World: A List of Valid Species Names*; Magnolia Press: Waco, TX, USA, 2010; Available online: http://hdl.handle.net/2263/17278 (accessed on 11 October 2020).

13. Latif, A.A.; Putterill, J.F.; De Klerk, D.G.; Pienaar, R.; Mans, B.J. *Nuttalliella namaqua* (*Ixodoidea*: *Nuttalliellidae*): First description of the male, immature stages and re-description of the female. *PLoS ONE* **2012**, *7*, e41651. [CrossRef] [PubMed]

14. Mans, B.J.; de Klerk, D.; Pienaar, R.; de Castro, M.H.; Latif, A.A. The mitochondrial genomes of *Nuttalliella namaqua* (*Ixodoidea*: *Nuttalliellidae*) and *Argas africolumbae* (*Ixodoidea*: *Argasidae*): Estimation of divergence dates for the major tick lineages and reconstruction of ancestral blood-feeding characters. *PLoS ONE* **2012**, *7*, e49461. [CrossRef] [PubMed]

15. Klemola, T.; Sormunen, J.J.; Mojzer, J.; Mäkelä, S.; Vesterinen, E.J. High tick abundance and diversity of tick-borne pathogens in a Finnish city. *Urban Ecosyst.* **2019**, *22*, 817–826. [CrossRef]

16. Kernif, T.; Leulmi, H.; Raoult, D.; Parola, P. Emerging tick-borne bacterial pathogens. *Microbiol. Spectr.* **2016**, *4*. [CrossRef] [PubMed]

17. Yusufmia, S.B.A.S.; Collins, N.E.; Nkuna, R.; Troskie, M.; Van den Bossche, P.; Penzhorn, B.L. Occurrence of *Theileria parva* and other haemoprotozoa in cattle at the edge of Hluhluwe-iMfolozi Park, KwaZulu-Natal, South Africa. *J. S. Afri. Vet. Assoc.* **2010**, *81*, 45–49. [CrossRef]

18. Jongejan, F.; Uilenberg, G. The global importance of ticks. *Parasitology* **2004**, *129*, S3–S14. [CrossRef] [PubMed]

19. Jensenius, M.; Parola, P.; Raoult, D. Threats to international travellers posed by tick-borne diseases. *Travel Med. Infect. Dis.* **2006**, *4*, 4–13. [CrossRef]

20. Hotez, P.J.; Bottazzi, M.E.; Strych, U.; Chang, L.Y.; Lim, Y.A.; Goodenow, M.M.; AbuBakar, S. Neglected tropical diseases among the Association of Southeast Asian Nations (ASEAN): Overview and update. *PLoS Negl. Trop. Dis.* **2015**, *9*, e0003575. [CrossRef]

21. Boularias, G.; Azzag, N.; Gandoin, C.; Bouillin, C.; Chomel, B.; Haddad, N.; Boulouis, H.J. Bovines Harbor a Diverse Array of Vector-Borne Pathogens in Northeast Algeria. *Pathogens* **2020**, *9*, 883. [CrossRef]

22. Rizzoli, A.; Silaghi, C.; Obiegala, A.; Rudolf, I.; Hubálek, Z.; Földvári, G.; Plantard, O.; Vayssier-Taussat, M.; Bonnet, S.; Špitalská, E.; et al. *Ixodes ricinus* and its transmitted pathogens in urban and peri-urban areas in Europe: New hazards and relevance for public health. *Front. Public Health* **2014**, *2*, 251. [CrossRef] [PubMed]

23. Sormunen, J.J.; Andersson, T.; Aspi, J.; Bäck, J.; Cederberg, T.; Haavisto, N.; Halonen, H.; Hänninen, J.; Inkinen, J.; Kulha, N.; et al. Monitoring of ticks and tick-borne pathogens through a nationwide research station network in Finland. *Ticks Tick-Borne Dis.* **2020**, *11*, 101449. [CrossRef]

24. Bilgic, H.B.; Bakırcı, S.; Kose, O.; Unlu, A.H.; Hacılarlıoglu, S.; Eren, H.; Weir, W.; Karagenc, T. Prevalence of tick-borne haemoparasites in small ruminants in Turkey and diagnostic sensitivity of single-PCR and RLB. *Parasites Vectors* **2017**, *10*, 211. [CrossRef] [PubMed]

25. Martínez-García, G.; Santamaría-Espinosa, R.M.; Lira-Amaya, J.J.; Figueroa, J.V. Challenges in Tick-Borne Pathogen Detection: The Case for *Babesia* spp. Identification in the Tick Vector. *Pathogens* **2021**, *10*, 92. [CrossRef] [PubMed]

26. Gitau, G.K.; Perry, B.D.; Katende, J.M.; McDermott, J.J.; Morzaria, S.P.; Young, A.S. The prevalence of serum antibodies to tick-borne infections in cattle in smallholder dairy farms in Murang'a District, Kenya: A cross-sectional study. *Prev. Vet. Med.* **1997**, *30*, 95–107. [CrossRef]

27. Hasle, G. Transport of ixodid ticks and tick-borne pathogens by migratory birds. *Front. Cell. Infect. Microbiol.* **2013**, *3*, 48. [CrossRef]

28. Makala, L.H.; Mangani, P.; Fujisaki, K.; Nagasawa, H. The current status of major tick borne diseases in Zambia. *Vet. Res.* **2003**, *34*, 27–45. [CrossRef]

29. Swai, E.S.; Karimuribo, E.D.; Kambarage, D.M.; Moshy, W.E.; Mbise, A.N. A comparison of seroprevalence and risk factors for *Theileria parva* and *T. mutans* in smallholder dairy cattle in the Tanga and Iringa regions of Tanzania. *Vet. J.* **2007**, *174*, 390–396. [CrossRef]

30. Barradas, P.F.; Mesquita, J.R.; Ferreira, P.; Gärtner, F.; Carvalho, M.; Inácio, E.; Chivinda, E.; Katimba, A.; Amorim, I. Molecular identification and characterization of *Rickettsia* spp. and other tick-borne pathogens in cattle and their ticks from Huambo, Angola. *Ticks Tick-Borne Dis.* **2021**, *12*, 101583. [CrossRef]

31. Kubelová, M.; Mazancová, J.; Široký, P. *Theileria, Babesia*, and *Anaplasma* detected by PCR in ruminant herds at Bié Province, Angola. *Parasite* **2012**, *19*, 417. [CrossRef]

32. Sili, G.; Byaruhanga, C.; Horak, I.; Steyn, H.; Chaisi, M.; Oosthuizen, M.C.; Neves, L. Ticks and tick-borne pathogens infecting livestock and dogs in Tchicala-Tcholoanga, Huambo Province, Angola. *Parasitol. Res.* **2021**, *120*, 1097–1102. [CrossRef] [PubMed]

33. Stoltsz, H.; Byaruhanga, C.; Troskie, M.; Makgabo, M.; Oosthuizen, M.C.; Collins, N.E.; Neves, L. Improved detection of *Babesia bigemina* from various geographical areas in Africa using quantitative PCR and reverse line blot hybridisation. *Ticks Tick-Borne Dis.* **2020**, *11*, 101415. [CrossRef] [PubMed]

34. Binta, M.G.; Losho, T.; Allsopp, B.A.; Mushi, E.Z. Isolation of *Theileria taurotragi* and *Theileria mutans* from cattle in Botswana. *Vet. Parasitol.* **1998**, *77*, 83–91. [CrossRef]

35. Berthelsson, J.; Ramabu, S.S.; Lysholm, S.; Aspán, A.; Wensman, J.J. *Anaplasma ovis* infection in goat flocks around Gaborone, Botswana. *Comp. Clin. Pathol.* **2020**, *29*, 167–172. [CrossRef]

36. Ramabu, S.S.; Kgwatalala, P.M.; Nsoso, S.J.; Gasebonwe, S.; Kgosiesele, E. *Anaplasma* infection prevalence in beef and dairy cattle in the southeast region of Botswana. *Vet. Parasitol. Reg. Stud. Rep.* **2017**, *12*, 4–8. [CrossRef]

37. Chatanga, E.; Kainga, H.; Maganga, E.; Hayashida, K.; Katakura, K.; Sugimoto, C.; Nonaka, N.; Nakao, R. Molecular identification and genetic characterization of tick-borne pathogens in sheep and goats at two farms in the central and southern regions of Malawi. *Ticks Tick-Borne Dis.* **2021**, *12*, 101629. [CrossRef]

38. De Jesus Fernandes, S.; Matos, C.A.; Freschi, C.R.; de Souza Ramos, I.A.; Machado, R.Z.; André, M.R. Diversity of *Anaplasma* species in cattle in Mozambique. *Ticks Tick-Borne Dis.* **2019**, *10*, 651–664. [CrossRef] [PubMed]

39. Martins, T.; Neves, L.; Pedro, O.; Fafetine, J.; Do Rosa-rio, V.; Domingos, A. Molecular detection of *Babesia* spp. and other haemoparasitic infections of cattle in Maputo Province, Mozambique. *Parasitology* **2010**, *137*, 939–946. [CrossRef]

40. Martins, T.M.; Pedro, O.C.; Caldeira, R.A.; do Rosário, V.E.; Neves, L.; Domingos, A. Detection of bovine babesiosis in Mozambique by a novel seminested hot-start PCR method. *Vet. Parasitol.* **2008**, *153*, 225–230. [CrossRef]

41. Matos, C.A.; Gonçalves, L.R.; de Souza Ramos, I.A.; Mendes, N.S.; Zanatto, D.C.S.; André, M.R.; Machado, R.Z. Molecular detection and characterization of *Ehrlichia ruminantium* from cattle in Mozambique. *Acta Trop.* **2019**, *191*, 198–203. [CrossRef]

42. Chaisi, M.E.; Baxter, J.R.; Hove, P.; Choopa, C.N.; Oosthuizen, M.C.; Brayton, K.A.; Khumalo, Z.T.; Mutshembele, A.M.; Mtshali, M.S.; Collins, N.E. Comparison of three nucleic acid-based tests for detecting *Anaplasma marginale* and *Anaplasma centrale* in cattle. *Onderstepoort J. Vet. Res.* **2017**, *84*, 1–9. [CrossRef]

43. Hove, P.; Khumalo, Z.T.; Chaisi, M.E.; Oosthuizen, M.C.; Brayton, K.A.; Collins, N.E. Detection and Characterisation of *Anaplasma marginale* and *A. centrale* in South Africa. *Vet. Sci.* **2018**, *5*, 26. [CrossRef]

44. Latif, A.A.; Troskie, P.C.; Peba, S.B.; Maboko, B.B.; Pienaar, R.; Mans, B.J. Corridor disease (buffalo-associated *Theileria parva*) outbreak in cattle introduced onto a game ranch and investigations into their carrier-state. *Vet. Parasitol. Reg. Stud. Rep.* **2019**, *18*, 100331. [CrossRef] [PubMed]

45. Marufu, M.C.; Chimonyo, M.; Mtshali, M.S.; Dzama, K.G. Molecular prevalence of *Anaplasma marginale* (*Rickettsiales*: *Anaplasmataceae*) in Nguni and local crossbred cattle under the low input production system in South Africa. In Proceedings of the 10th Annual Congress of the Southern African Society for Veterinary Epidemiology and Preventive Medicine, Farm Inn, South Africa, 1–3 August 2012; p. 57.

46. Mbizeni, S.; Potgieter, F.T.; Troskie, C.; Mans, B.J.; Penzhorn, B.L.; Latif, A.A. Field and laboratory studies on Corridor disease (Theileria parva infection) in cattle population at the livestock/game interface of uPhongolo-Mkuze area, South Africa. *Ticks Tick-Borne Dis.* **2013**, *4*, 227–234. [CrossRef] [PubMed]

47. Mofokeng, L.S.; Taioe, O.M.; Smit, N.J.; Thekisoe, O.M. Parasites of veterinary importance from domestic animals in uMkhanyakude district of KwaZulu-Natal province. *J. S. Afr. Vet. Assoc.* **2020**, *91*, 1–11. [CrossRef] [PubMed]

48. Mtshali, M.S.; Mtshali, P.S. Molecular diagnosis and phylogenetic analysis of *Babesia bigemina* and *Babesia bovis* hemoparasites from cattle in South Africa. *BMC Vet. Res.* **2013**, *9*, 154. [CrossRef]

49. Mtshali, P.S.; Mtshali, M.S. In silico and phylogenetic analyses of partial BbRAP-1, BbCP2, BbSBP-4 and BbβTUB gene sequences of *Babesia bovis* isolates from cattle in South Africa. *BMC Vet. Res.* **2017**, *13*, 383. [CrossRef] [PubMed]

50. Mtshali, M.S.; De la Fuente, J.; Ruybal, P.; Kocan, K.M.; Vicente, J.; Mbati, P.A.; Shkap, V.; Blouin, E.F.; Mohale, N.E.; Moloi, T.P.; et al. Prevalence and genetic diversity of *Anaplasma marginale* strains in cattle in South Africa. *Zoonoses Public Health* **2007**, *54*, 23–30. [CrossRef]

51. Mtshali, M.S.; Steyn, H.C.; Mtshali, P.S.; Mbati, P.A.; Kocan, K.M.; Latif, A.; Shkap, V. The detection and characterization of multiple tick-borne pathogens in cattle at Ficksburg and Reitz (Free State Province, South Africa) using reverse line blot hybridization. *Afri. J. Microbiol. Res.* **2013**, *7*, 646–651. [CrossRef]

52. Mtshali, P.S.; Tsotetsi, A.M.; Thekisoe, M.M.O.; Mtshali, M.S. Nested PCR detection and phylogenetic analysis of *Babesia bovis* and *Babesia bigemina* in cattle from peri-urban localities in Gauteng province, South Africa. *J. Vet. Med. Sci.* **2013**, *76*, 145–150. [CrossRef]

53. Mutshembele, A.M.; Cabezas-Cruz, A.; Mtshali, M.S.; Thekisoe, O.M.; Galindo, R.C.; de la Fuente, J. Epidemiology and evolution of the genetic variability of *Anaplasma marginale* in South Africa. *Ticks Tick-Borne Dis.* **2014**, *5*, 624–631. [CrossRef] [PubMed]

54. Pienaar, R.; Troskie, P.C.; Josemans, A.I.; Potgieter, F.T.; Maboko, B.B.; Latif, A.A.; Mans, B.J. Investigations into the carrier-state of *Theileria* spp.(buffalo) in cattle. *Int. J. Parasitol. Parasites Wildl.* **2002**, *11*, 136–142. [CrossRef] [PubMed]

55. Ringo, A.E.; Moumouni, P.F.A.; Taioe, M.; Jirapattharasate, C.; Liu, M.; Wang, G.; Gao, Y.; Guo, H.; Lee, S.H.; Zheng, W.; et al. Molecular analysis of tick-borne protozoan and rickettsial pathogens in small ruminants from two South African provinces. *Parasitol. Int.* **2018**, *67*, 144–149. [CrossRef] [PubMed]

56. Steyn, H.C.; Pretorius, A. Genetic diversity of *Ehrlichia ruminantium* field strains from selected farms in South Africa. *Onderstepoort J. Vet. Res.* **2020**, *87*, 1–12. [CrossRef] [PubMed]

57. Thompson, B.E.; Latif, A.A.; Oosthuizen, M.C.; Troskie, M.; Penzhorn, B.L. Occurrence of *Theileria parva* infection in cattle on a farm in the Ladysmith district, KwaZulu-Natal, South Africa. *J. S. Afri. Vet. Assoc.* **2008**, *79*, 31–35. [CrossRef]

58. Laisser, E.L.K.; Kipanyula, M.J.; Msalya, G.; Mdegela, R.H.; Karimuribo, E.D.; Mwilawa, A.J.; Mwega, E.D.; Kusiluka, L.; Chenyambuga, S.W. Tick burden and prevalence of *Theileria parva* infection in Tarime zebu cattle in the lake zone of Tanzania. *Trop. Anim. Health Prod.* **2014**, *46*, 1391–1396. [CrossRef]

59. Kazungu, Y.E.; Mwega, E.; Neselle, M.O.; Sallu, R.; Kimera, S.I.; Gwakisa, P. Incremental effect of natural tick challenge on the infection and treatment method-induced immunity against *T. parva* in cattle under agro-pastoral systems in Northern Tanzania. *Ticks Tick-Borne Dis.* **2015**, *6*, 587–591. [CrossRef]

60. Kerario, I.I.; Chenyambuga, S.W.; Mwega, E.D.; Rukambile, E.; Simulundu, E.; Simuunza, M.C. Diversity of two *Theileria parva* CD8+ antigens in cattle and buffalo-derived parasites in Tanzania. *Ticks Tick-Borne Dis.* **2019**, *10*, 1003–1017. [CrossRef]

61. Kimaro, E.G.; Mor, S.M.; Gwakisa, P.; Toribio, J.A. Seasonal occurrence of *Theileria parva* infection and management practices amongst Maasai pastoralist communities in Monduli District, Northern Tanzania. *Vet. Parasitol.* **2017**, *246*, 43–52. [CrossRef]

62. Magulu, E.; Kindoro, F.; Mwega, E.; Kimera, S.; Shirima, G.; Gwakisa, P. Detection of carrier state and genetic diversity of *Theileria parva* in ECF-vaccinated and naturally exposed cattle in Tanzania. *Vet. Parasitol. Reg. Stud. Rep.* **2019**, *17*, 100312. [CrossRef]

63. Mwamuye, M.M.; Odongo, D.; Kazungu, Y.; Kindoro, F.; Gwakisa, P.; Bishop, R.P.; Nijhof, A.M.; Obara, I. Variant analysis of the sporozoite surface antigen gene reveals that asymptomatic cattle from wildlife-livestock interface areas in northern Tanzania harbour buffalo-derived *T. parva*. *Parasitol. Res.* **2020**, *119*, 3817–3828. [CrossRef]

64. Ringo, A.E.; Moumouni, P.F.A.; Lee, S.H.; Liu, M.; Khamis, Y.H.; Gao, Y.; Guo, H.; Zheng, W.; Efstratiou, A.; Galon, E.M.; et al. Molecular detection and characterization of tick-borne protozoan and rickettsial pathogens isolated from cattle on Pemba Island, Tanzania. *Ticks Tick-Borne Dis.* **2018**, *9*, 1437–1445. [CrossRef] [PubMed]

65. Ringo, A.E.; Rizk, M.A.; Moumouni, P.F.A.; Liu, M.; Galon, E.M.; Li, Y.; Ji, S.; Tumwebaze, M.; Byamukama, B.; Thekisoe, O.; et al. Molecular detection and characterization of tick-borne haemoparasites among cattle on Zanzibar Island, Tanzania. *Acta Trop.* **2020**, *211*, 105598. [CrossRef]

66. Rukambile, E.; Machuka, E.; Njahira, M.; Kyalo, M.; Skilton, R.; Mwega, E.; Chota, A.; Mathias, M.; Sallu, R.; Salih, D. Population genetic analysis of *Theileria parva* isolated in cattle and buffaloes in Tanzania using minisatellite and microsatellite markers. *Vet. Parasitol.* **2016**, *224*, 20–26. [CrossRef] [PubMed]

67. Thekisoe, O.M.M.; Omolo, J.D.; Swai, E.S.; Hayashida, K.; Zhang, J.; Sugimoto, C.; Inoue, N. Preliminary application and evaluation of loop-mediated isothermal amplification (LAMP) for detection of bovine theileriosis and trypanosomosis in Tanzania: Research communication. *Onderstepoort J. Vet. Res.* **2007**, *74*, 339–342. [CrossRef] [PubMed]

68. Iseki, H.; Alhassan, A.; Ohta, N.; Thekisoe, O.M.; Yokoyama, N.; Inoue, N.; Nambota, A.; Yasuda, J.; Igarashi, I. Development of a multiplex loop-mediated isothermal amplification (mLAMP) method for the simultaneous detection of bovine *Babesia* parasites. *J. Microbial. Methods* **2007**, *71*, 281–287. [CrossRef]

69. Muleya, W.; Namangala, B.; Simuunza, M.; Nakao, R.; Inoue, N.; Kimura, T.; Ito, K.; Sugimoto, C.; Sawa, H. Population genetic analysis and sub-structuring of *Theileria parva* in the northern and eastern parts of Zambia. *Parasites Vectors* **2012**, *5*, 255. [CrossRef] [PubMed]

70. Musinguzi, S.P.; Suganuma, K.; Asada, M.; Laohasinnarong, D.; Sivakumar, T.; Yokoyama, N.; Namangala, B.; Sugimoto, C.; Suzuki, Y.; Xuan, X.; et al. A PCR-based survey of animal African trypanosomosis and selected piroplasm parasites of cattle and goats in Zambia. *J. Vet. Med. Sci.* **2016**, *78*, 1819–1824. [CrossRef] [PubMed]

71. Simuunza, M.; Weir, W.; Courcier, E.; Tait, A.; Shiels, B. Epidemiological analysis of tick-borne diseases in Zambia. *Vet. Parasitol.* **2011**, *175*, 331–342. [CrossRef]

72. Squarre, D.; Nakamura, Y.; Hayashida, K.; Kawai, N.; Chambaro, H.; Namangala, B.; Sugimoto, C.; Yamagishi, J. Investigation of the piroplasm diversity circulating in wildlife and cattle of the greater Kafue ecosystem, Zambia. *Parasites Vectors* **2020**, *13*, 599. [CrossRef]

73. Tembo, S.; Collins, N.E.; Sibeko-Matjila, K.P.; Troskie, M.; Vorster, I.; Byaruhanga, C.; Oosthuizen, M.C. Occurrence of tick-borne haemoparasites in cattle in the Mungwi District, Northern Province, Zambia. *Ticks Tick-Borne Dis.* **2018**, *9*, 707–717. [CrossRef]

74. Yamada, S.; Konnai, S.; Imamura, S.; Simuunza, M.; Chembensofu, M.; Chota, A.; Nambota, A.; Onuma, M.; Ohashi, K. PCR-based detection of blood parasites in cattle and adult *Rhipicephalus appendiculatus* ticks. *Vet. J.* **2009**, *182*, 352–355. [CrossRef] [PubMed]

75. Smeenk, I.; Kelly, P.J.; Wray, K.; Musuka, G.; Trees, A.J.; Jongejan, F. *Babesia bovis* and *B. bigemina* DNA detected in cattle and ticks from Zimbabwe by polymerase chain reaction. *J. S. Afri. Vet. Assoc.* **2000**, *71*, 21–24. [CrossRef] [PubMed]

76. Yssouf, A.; Socolovschi, C.; Kernif, T.; Temmam, S.; Lagadec, E.; Tortosa, P.; Parola, P. First molecular detection of *Rickettsia africae* in ticks from the Union of the Comoros. *Parasites Vectors* **2014**, *7*, 444. [CrossRef] [PubMed]

77. Ehlers, J.; Krüger, A.; Rakotondranary, S.J.; Ratovonamana, R.Y.; Poppert, S.; Ganzhorn, J.U.; Tappe, D. Molecular detection of *Rickettsia* spp., *Borrelia* spp., *Bartonella* spp. and *Yersinia pestis* in ectoparasites of endemic and domestic animals in southwest Madagascar. *Acta Trop.* **2020**, *205*, 105339. [CrossRef] [PubMed]

78. Pothmann, D.; Poppert, S.; Rakotozandrindrainy, R.; Hogan, B.; Mastropaolo, M.; Thiel, C.; Silaghi, C. Prevalence and genetic characterization of *Anaplasma marginale* in zebu cattle (*Bos indicus*) and their ticks (*Amblyomma variegatum*, *Rhipicephalus microplus*) from Madagascar. *Ticks Tick-Borne Dis.* **2016**, *7*, 1116–1123. [CrossRef]

79. Matsimbe, A.M.; Magaia, V.; Sanches, G.S.; Neves, L.; Noormahomed, E.; Antunes, S.; Domingos, A. Molecular detection of pathogens in ticks infesting cattle in Nampula province, Mozambique. *Exp. Appl. Acarol.* **2017**, *73*, 91–102. [CrossRef]

80. Adelabu, O.A.; Iweriebor, B.C.; Okoh, A.I.; Obi, L.C. Phylogenetic profiling for zoonotic *Ehrlichia* spp. from ixodid ticks in the Eastern Cape, South Africa. *Transbound. Emerg. Dis.* **2020**, *67*, 1247–1256. [CrossRef]

81. Berggoetz, M.; Schmid, M.; Ston, D.; Wyss, V.; Chevillon, C.; Pretorius, A.M.; Gern, L. Protozoan and bacterial pathogens in tick salivary glands in wild and domestic animal environments in South Africa. *Ticks Tick-Borne Dis.* **2014**, *5*, 176–185. [CrossRef]

82. Guo, H.; Moumouni, P.F.A.; Thekisoe, O.; Gao, Y.; Liu, M.; Li, J.; Galon, E.M.; Efstratiou, A.; Wang, G.; Jirapattharasate, C.; et al. Genetic characterization of tick-borne pathogens in ticks infesting cattle and sheep from three South African provinces. *Ticks Tick-Borne Dis.* **2019**, *10*, 875–882. [CrossRef]

83. Iweriebor, B.C.; Mmbaga, E.J.; Adegborioye, A.; Igwaran, A.; Obi, L.C.; Okoh, A.I. Genetic profiling for *Anaplasma* and *Ehrlichia* species in ticks collected in the Eastern Cape Province of South Africa. *BMC Microbial.* **2017**, *17*, 45. [CrossRef] [PubMed]

84. Iweriebor, B.C.; Nqoro, A.; Obi, C.L. *Rickettsia africae* an Agent of African Tick Bite Fever in Ticks Collected from Domestic Animals in Eastern Cape, South Africa. *Pathogens* **2020**, *9*, 631. [CrossRef] [PubMed]

85. Kolo, A.O.; Collins, N.E.; Brayton, K.A.; Chaisi, M.; Blumberg, L.; Frean, J.; Gall, C.A.; MWentzel, J.; Wills-Berriman, S.; Boni, L.D.; et al. *Anaplasma phagocytophilum* and Other *Anaplasma* spp. in Various Hosts in the Mnisi Community, Mpumalanga Province, South Africa. *Microorganisms* **2020**, *8*, 1812. [CrossRef]

86. Jongejan, F.; Berger, L.; Busser, S.; Deetman, I.; Jochems, M.; Leenders, T.; De Sitter, B.; Van der Steen, F.; Wentzel, J.; Stoltsz, H. *Amblyomma hebraeum* is the predominant tick species on goats in the Mnisi Community Area of Mpumalanga Province South Africa and is co-infected with *Ehrlichia ruminantium* and *Rickettsia africae*. *Parasites Vectors* **2020**, *13*, 172. [CrossRef]

87. Damian, D.; Damas, M.; Wensman, J.J.; Berg, M. Molecular Diversity of Hard Tick Species from Selected Areas of a Wildlife-Livestock Interface Ecosystem at Mikumi National Park, Morogoro Region, Tanzania. *Vet. Sci.* **2021**, *8*, 36. [CrossRef] [PubMed]

88. Fyumagwa, R.D.; Simmler, P.; Meli, M.L.; Hoare, R.; Hofmann-Lehmann, R.; Lutz, H. Prevalence of *Anaplasma marginale* in different tick species from Ngorongoro Crater, Tanzania. *Vet. Parasitol.* **2009**, *161*, 154–157. [CrossRef]

89. Kim, T.Y.; Kwak, Y.S.; Kim, J.Y.; Nam, S.H.; Lee, I.Y.; Mduma, S.; Keyyu, J.; Fyumagwa, R.; Yong, T.S. Prevalence of tick-borne pathogens from ticks collected from cattle and wild animals in Tanzania in 2012. *Korean J. Parasitol.* **2018**, *56*, 305. [CrossRef] [PubMed]

90. Savadye, D.T.; Kelly, P.J.; Mahan, S.M. Evidence to show that an agent that cross-reacts serologically with *Cowdria ruminantium* in Zimbabwe is transmitted by ticks. *Exp. Appl. Acarol.* **1998**, *22*, 111–122. [CrossRef]

91. Gilbert, L. Louping ill virus in the UK: A review of the hosts, transmission and ecological consequences of control. *Exp. Appl. Acarol.* **2016**, *68*, 363–374. [CrossRef]

92. Randolph, S.E. Tick ecology: Processes and patterns behind the epidemiological risk posed by ixodid ticks as vectors. *Parasitology* **2004**, *129*, S37–S65. [CrossRef]

93. Njiiri, E.N. The Occurrence of *Ehrlichia ruminantium* and Other Haemoparasites in Calves in Western Kenya Determined by Reverse Line Blot Hybridization Assay, Real-Time PCR and Nested PCR. Ph.D. Thesis, University of Pretoria, Pretoria, South Africa, 2013. Available online: http://hdl.handle.net/2263/26197 (accessed on 22 January 2021).

94. Basu, A.K.; Charles, R. Chapter 1—A general account of ticks. In *Ticks of Trinidad and Tobago—An Overview*; Academic Press: Cambridge, MA, USA, 2017; pp. 1–33.

95. Ghafar, A.; Abbas, T.; Rehman, A.; Sandhu, Z.U.D.; Cabezas-Cruz, A.; Jabbar, A. Systematic review of ticks and tick-borne pathogens of small ruminants in Pakistan. *Pathogens* **2020**, *9*, 937. [CrossRef] [PubMed]

96. Macaluso, K.R.; Davis, J.O.N.; Alam, U.; Korman, A.M.Y.; Rutherford, J.S.; Rosenberg, R.; Azad, A.F. Spotted fever group rickettsiae in ticks from the Masai Mara region of Kenya. *Am. J. Trop. Med. Hyg.* **2003**, *68*, 551–553. [CrossRef] [PubMed]

97. Mediannikov, O.; Diatta, G.; Zolia, Y.; Balde, M.C.; Kohar, H.; Trape, J.F.; Raoult, D. Tick-borne rickettsiae in Guinea and Liberia. *Ticks Tick-Borne Dis.* **2012**, *3*, 43–48. [CrossRef]

98. Hornok, S.; Abichu, G.; Meli, M.L.; Tánczos, B.; Sulyok, K.M.; Gyuranecz, M.; Gönczi, E.; Farkas, R.; Hofmann-Lehmann, R. Influence of the biotope on the tick infestation of cattle and on the tick-borne pathogen repertoire of cattle ticks in Ethiopia. *PLoS ONE* **2014**, *9*, e106452. [CrossRef] [PubMed]

99. Kumsa, B.; Socolovschi, C.; Almeras, L.; Raoult, D.; Parola, P. Occurrence and genotyping of *Coxiella burnetii* in ixodid ticks in Oromia, Ethiopia. *Am. J. Trop. Med. Hyg.* **2015**, *93*, 1074. [CrossRef]

100. Gondard, M.; Cabezas-Cruz, A.; Charles, R.A.; Vayssier-Taussat, M.; Albina, E.; Moutailler, S. Ticks and tick-borne pathogens of the Caribbean: Current understanding and future directions for more comprehensive surveillance. *Front. Cell. Infect. Microbial.* **2017**, *7*, 490. [CrossRef] [PubMed]

101. Aktas, M.; Altay, K.; Ozubek, S.; Dumanli, N. A survey of ixodid ticks feeding on cattle and prevalence of tick-borne pathogens in the Black Sea region of Turkey. *Vet. Parasitol.* **2012**, *187*, 567–571. [CrossRef]

102. Sivakumar, T.; Tattiyapong, M.; Okubo, K.; Suganuma, K.; Hayashida, K.; Igarashi, I.; Zakimi, S.; Matsumoto, K.; Inokuma, H.; Yokoyama, N. PCR detection of *Babesia ovata* from questing ticks in Japan. *Ticks Tick-Borne Dis.* **2014**, *5*, 305–310. [CrossRef]

103. Chaligiannis, I.; de Mera, I.G.F.; Papa, A.; Sotiraki, S.; de la Fuente, J. Molecular identification of tick-borne pathogens in ticks collected from dogs and small ruminants from Greece. *Exp. Appl. Acarol.* **2018**, *74*, 443–453. [CrossRef]

104. Mossaad, E.; Gaithuma, A.; Mohamed, Y.O.; Suganuma, K.; Umemiya-Shirafuji, R.; Ohari, Y.; Salim, B.; Liu, M.; Xuan, X. Molecular Characterization of Ticks and Tick-Borne Pathogens in Cattle from Khartoum State and East Darfur State, Sudan. *Pathogens* **2021**, *10*, 580. [CrossRef]

105. Rehman, A.; Conraths, F.J.; Sauter-Louis, C.; Krücken, J.; Nijhof, A.M. Epidemiology of tick-borne pathogens in the semi-arid and the arid agro-ecological zones of Punjab province, Pakistan. *Transbound. Emerg. Dis.* **2019**, *66*, 526–536. [CrossRef] [PubMed]

106. Raoult, D.; Roux, V. Rickettsioses as paradigms of new or emerging infectious diseases. *Clin. Microbiol. Rev.* **1997**, *10*, 694–719. [CrossRef] [PubMed]

107. Magaia, V.; Taviani, E.; Cangi, N.; Neves, L. Molecular detection of *Rickettsia africae* in *Amblyomma* ticks collected in cattle from Southern and Central Mozambique. *J. Infect. Dev. Ctries.* **2020**, *14*, 614–622. [CrossRef]

108. Althaus, F.; Greub, G.; Raoult, D.; Genton, B. African tick-bite fever: A new entity in the differential diagnosis of multiple eschars in travelers. Description of five cases imported from South Africa to Switzerland. *Int. J. Infect. Dis.* **2010**, *14*, e274–e276. [CrossRef]

109. Sadeddine, R.; Diarra, A.Z.; Laroche, M.; Mediannikov, O.; Righi, S.; Benakhla, A.; Dahmana, H.; Raoult, D.; Parola, P. Molecular identification of protozoal and bacterial organisms in domestic animals and their infesting ticks from north-eastern Algeria. *Ticks Tick-Borne Dis.* **2020**, *11*, 101330. [CrossRef]

110. Byamukama, B.; Vudriko, P.; Tumwebaze, M.A.; Tayebwa, D.S.; Byaruhanga, J.; Angwe, M.K.; LI, J.; Galon, E.M.; Ringo, A.; Liu, M.; et al. Molecular detection of selected tick-borne pathogens infecting cattle at the wildlife–livestock interface of Queen Elizabeth National Park in Kasese District, Uganda. *Ticks Tick-Borne Dis.* **2021**, *12*, 101772. [CrossRef]

111. Belkahia, H.; Said, M.B.; El Hamdi, S.; Yahiaoui, M.; Gharbi, M.; Daaloul-Jedidi, M.; Mhadhbi, M.; Jedidi, M.; Darghouth, M.A.; Klabi, I.; et al. First molecular identification and genetic characterization of *Anaplasma ovis* in sheep from Tunisia. *Small Rumin. Res.* **2014**, *121*, 404–410. [CrossRef]

112. Estrada-Peña, A.; Santos-Silva, M.M. The distribution of ticks (*Acari*: *Ixodidae*) of domestic livestock in Portugal. *Exp. Appl. Acarol.* **2005**, *36*, 233–246. [CrossRef]

113. Zhang, J.; Kelly, P.; Guo, W.; Xu, C.; Wei, L.; Jongejan, F.; Loftis, A.; Wang, C. Development of a generic *Ehrlichia* FRET-qPCR and investigation of ehrlichioses in domestic ruminants on five Caribbean islands. *Parasites Vectors* **2015**, *8*, 506. [CrossRef]

114. Bell-Sakyi, L.; Koney, E.B.M.; Dogbey, O.; Walker, A.R. Incidence and prevalence of tick-borne haemoparasites in domestic ruminants in Ghana. *Vet. Parasitol.* **2004**, *124*, 25–42. [CrossRef]

115. Southern African Development Community (SADC). Livestock Production. 2012. Available online: https://www.sadc.int/themes/agriculture-food-security/livestock-production/ (accessed on 5 July 2021).

116. Muhanguzi, D.; Ikwap, K.; Picozzi, K.; Waiswa, C. Molecular characterization of *Anaplasma* and *Ehrlichia* species in different cattle breeds and age groups in Mbarara district (Western Uganda). *Int. J. Anim. Vet. Adv.* **2010**, *2*, 76–88.

117. De la Fuente, J.; Torina, A.; Caracappa, S.; Tumino, G.; Furlá, R.; Almazán, C.; Kocan, K.M. Serologic and molecular characterization of *Anaplasma* species infection in farm animals and ticks from Sicily. *Vet. Parasitol.* **2005**, *133*, 357–362. [CrossRef] [PubMed]

118. Noaman, V.; Shayan, P.; Amininia, N. Molecular diagnostic of *Anaplasma marginale* in carrier cattle. *Iran. J. Parasitol.* **2009**, *4*, 26–33.

119. Aktas, M.; Altay, K.; Dumanli, N. Molecular detection and identification of *Anaplasma* and *Ehrlichia* species in cattle from Turkey. *Ticks Tick-Borne Dis.* **2011**, *2*, 62–65. [CrossRef]

120. Soosaraei, M.; Haghi, M.M.; Etemadifar, F.; Fakhar, M.; Teshnizi, S.H.; Asfaram, S.; Esboei, B.R. Status of *Anaplasma* spp. infection in domestic ruminants from Iran: A systematic review with meta-analysis. *Parasite Epidemiol. Control* **2020**, *11*, e00173. [CrossRef]

121. Mohammadian, B.; Noaman, V.; Emami, S.J. Molecular survey on prevalence and risk factors of *Anaplasma* spp. infection in cattle and sheep in West of Iran. *Trop. Anim. Health Prod.* **2021**, *53*, 266. [CrossRef]

122. Salehi-Guilandeh, S.; Sadeghi-Dehkordi, Z.; Sadeghi-Nasab, A. Molecular detection of *Anaplasma* spp. in cattle of Talesh County, North of Iran. Bulg. *J. Vet. Med.* **2019**, *22*, 457–465. [CrossRef]

123. Liu, X.; Yan, B.; Wang, Q.; Jiang, M.; Tu, C.; Chen, C.; Hornok, S.; Wang, Y. *Babesia vesperuginis* in common pipistrelle (*Pipistrellus pipistrellus)* and the bat soft tick *Argas vespertilionis* in the People's Republic of China. *J. Wildl. Dis.* **2018**, *54*, 419–421. [CrossRef]

124. Amorim, L.S.; Wenceslau, A.A.; Carvalho, F.S.; Carneiro, P.L.S.; Albuquerque, G.R. Bovine babesiosis and anaplasmosis complex: Diagnosis and evaluation of the risk factors from Bahia, Brazil. *Rev. Bras. Parasitol. Vet.* **2014**, *23*, 328–336. [CrossRef]

125. Jaimes-Dueñez, J.; Triana-Chávez, O.; Holguín-Rocha, A.; Tobon-Castaño, A.; Mejía-Jaramillo, A.M. Molecular surveillance and phylogenetic traits of *Babesia bigemina* and *Babesia bovis* in cattle (*Bos taurus*) and water buffaloes (*Bubalus bubalis*) from Colombia. *Parasites Vectors* **2018**, *11*, 510. [CrossRef]

126. Kocan, K.M. Targeting ticks for control of selected hemoparasitic diseases of cattle. *Vet. Parasitol.* **1995**, *57*, 121–151. [CrossRef]

127. Esemu, S.N.; Ndip, R.N.; Ndip, L.M. Detection of *Ehrlichia ruminantium* infection in cattle in Cameroon. *BMC Res. Notes* **2018**, *11*, 217. [CrossRef] [PubMed]

128. Hailemariam, Z.; Krücken, J.; Baumann, M.; Ahmed, J.S.; Clausen, P.H.; Nijhof, A.M. Molecular detection of tick-borne pathogens in cattle from Southwestern Ethiopia. *PLoS ONE* **2017**, *12*, e0188248. [CrossRef] [PubMed]

129. Lorusso, V.; Wijnveld, M.; Majekodunmi, A.O.; Dongkum, C.; Fajinmi, A.; Dogo, A.G.; Thrusfield, M.; Mugenyi, A.; Vaumourin, E.; Igweh, A.C.; et al. Tick-borne pathogens of zoonotic and veterinary importance in Nigerian cattle. *Parasites Vectors* **2016**, *9*, 388. [CrossRef]

130. Deem, S.L.; Noval, R.A.I.; Yonow, T.; Peter, T.F.; Mahan, S.M.; Burridge, M.J. The epidemiology of heartwater: Establishment and maintenance of endemic stability. *Parasitol. Today* **1996**, *12*, 402–405. [CrossRef]

131. Leeflang, P.; Ilemobade, A.A. 1977. Tick-borne diseases of domestic animals in Northern Nigeria. *Trop. Anim. Health Prod.* **1977**, *9*, 211–218. [CrossRef]

132. Moumouni, P.F.A.; Aboge, G.O.; Terkawi, M.A.; Masatani, T.; Cao, S.; Kamyingkird, K.; Jirapattharasate, C.; Zhou, M.; Wang, G.; Liu, M.; et al. Molecular detection and characterization of *Babesia bovis, Babesia bigemina, Theileria* species and *Anaplasma marginale* isolated from cattle in Kenya. *Parasites Vectors* **2015**, *8*, 496. [CrossRef] [PubMed]

133. Oura, C.A.; Tait, A.; Asiimwe, B.; Lubega, G.W.; Weir, W. *Theileria parva* genetic diversity and haemoparasite prevalence in cattle and wildlife in and around Lake Mburo National Park in Uganda. *Parasitol. Res.* **2011**, *108*, 1365–1374. [CrossRef]

134. Salih, D.A.; El Hussein, A.M.; Kyule, M.N.; Zessin, K.H.; Ahmed, J.S.; Seitzer, U. Determination of potential risk factors associated with *Theileria annulata* and *Theileria parva* infections of cattle in the Sudan. *Parasitol. Res.* **2007**, *101*, 1285–1288. [CrossRef]

135. Tomassone, L.; Grego, E.; Callà, G.; Rodighiero, P.; Pressi, G.; Gebre, S.; Zeleke, B.; De Meneghi, D. Ticks and tick-borne pathogens in livestock from nomadic herds in the Somali Region, Ethiopia. *Exp. Appl. Acarol.* **2012**, *56*, 391–401. [CrossRef]

136. Byaruhanga, C.; Collins, N.E.; Knobel, D.; Chaisi, M.E.; Vorster, I.; Steyn, H.C.; Oosthuizen, M.C. Molecular investigation of tick-borne haemoparasite infections among transhumant zebu cattle in Karamoja Region, Uganda. *Vet. Parasitoi. Reg. Stud. Rep.* **2016**, *3*, 27–35. [CrossRef] [PubMed]

137. Okal, M.N.; Odhiambo, B.K.; Otieno, P.; Bargul, J.L.; Masiga, D.; Villinger, J.; Kalayou, S. *Anaplasma* and *Theileria* pathogens in Cattle of Lambwe Valley, Kenya: A Case for Pro-Active Surveillance in the Wildlife–Livestock Interface. *Microorganisms* **2020**, *8*, 1830. [CrossRef] [PubMed]

138. Oura, C.A.L.; Bishop, R.P.; Wampande, E.M.; Lubega, G.W.; Tait, A. Application of a reverse line blot assay to the study of haemoparasites in cattle in Uganda. *Int. J. Parasitol.* **2004**, *34*, 603–613. [CrossRef] [PubMed]

139. Schnittger, L.; Yin, H.; Qi, B.; Gubbels, M.J.; Beyer, D.; Niemann, S.; Jongejan, F.; Ahmed, J.S. Simultaneous detection and differentiation of *Theileria* and *Babesia* parasites infecting small ruminants by reverse line blotting. *Parasitol. Res.* **2004**, *92*, 189–196. [CrossRef] [PubMed]

140. Borenstein, M.; Rothstein, H.; Cohen, J. *Comprehensive Meta-Analysis: A Computer Program for Research Synthesis*; Biostat: Englewood, NJ, USA, 1999.

141. Higgins, J.P.; Thompson, S.G.; Deeks, J.J.; Altman, D.G. Measuring inconsistency in meta-analyses. *BMJ* **2003**, *327*, 557–560. [CrossRef] [PubMed]

142. Begg, C.B.; Mazumdar, M. Operating characteristics of a rank correlation test for publication bias. *Biometrics* **1994**, *50*, 1088–1101. [CrossRef] [PubMed]