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Supplementary Table 1. Locality and countries from which stocks of *Rhipicephalus microplus*, *Amblyomma variegatum* and *Rhipicephalus appendiculatus* were collected

Locality, Country	<i>R. microplus</i>	<i>A. variegatum</i>	<i>R. appendiculatus</i>
East Africa			
Madibila, Tanzania	x	x	x
Chamakweza, Tanzania	x	x	x
Serere, Uganda	x	x	x
Serere 1, Uganda	x		x
Serere 2, Uganda			x
Oromia, Ethiopia		x	
West Africa			
Donga, Benin	x	x	
Zou, Benin	x		
Akuse, Ghana	x	x	
Narth Korkpe, Ghana	x		
Quanpam, Nigeria	x	x	
Soba, Nigeria	x		

Supplementary Table 2. Summary of tick infestations and tick counts

Study no.	(No. ticks) and infestation days	Assessment days
Exploratory studies		
385	Cattle were infested with approximately 3 000 <i>R. microplus</i> larvae per bovine on Days -28, -25, -23, -21, -18, -15, -14, -10, -8 and -4 for the purposes of randomisation and therapeutic efficacy assessments and approximately 5 000 <i>R. microplus</i> larvae) Days 21, 25, 28, 49, 52, 56, 77, 80 and 84 for persistent efficacy assessments.	Therapeutic efficacy: fully engorged female ticks collected and counted from Days 1 to 21. Persistent efficacy: engorged female ticks were collected and counted from Days 42 to 56 (all groups) Days 71 to 84, Days 99 to 112 (groups 1 to 3).
387	Cattle were infested with approximately 3 000 <i>R. microplus</i> larvae applied per bovine on Days -28, -25, -23, -20, -18, -15, -13, -11 -8 and -4 for the purposes of randomisation and therapeutic efficacy assessments and approximately 5 000 <i>R. microplus</i> larvae) Days 21, 24, 28, 49, 52, 56, 77 and 80 for persistent efficacy assessments.	Therapeutic efficacy: fully engorged female ticks were collected and counted from Days 1 to 21. Persistent efficacy: fully engorged female ticks were collected and counted from Days 42 to 56 for all groups and Days 70 to 83 for groups 2 and 4.
391	Approximately 30 adult unfed <i>A. variegatum</i> ticks (± 15 males and ± 15 females) were applied per goat on Day -14 for randomisation purposes and Day -4 for evaluation of therapeutic efficacy. Goats were reinfested (approximately 20 adult unfed <i>A. variegatum</i> ticks (± 10 males and ± 10 females)) on Days 14 and 28 to assess persistent efficacy in groups 2 and 3 and therapeutic efficacy in group 4. Group 3 were also infested on Day 42 to assess persistent efficacy, Group 4 were infested on Days 42 and 56 to assess therapeutic efficacy	Tick counts were performed at 48, 72 and 96 h (± 4 h) after infestation or treatment administration.
389	30 adult, unfed <i>R. appendiculatus</i> ticks per ear) applied per bovine on Day -6 for randomisation purposes, on Day -1 for evaluation of therapeutic efficacy, Days 14, 28, 56 and 63 to assess persistent efficacy for groups 2 and 3, and therapeutic efficacy for the respective treatments in group 4.	In-situ tick counts were conducted on Day 1 (groups 1 to 3) and Day 65 (all groups). Ticks were counted and removed on Days -4, 2, 16, 30, 58 ($48 \text{ h} \pm 4 \text{ h}$ after tick infestation or treatment administration for groups 2 and 3, or $24 \text{ h} \pm 4 \text{ h}$ after treatment administration for group 4) and 66 ($72 \text{ h} \pm 4 \text{ h}$ after tick infestation).
Confirmatory studies		
386	Cattle were infested with approximately 3 000 <i>R. microplus</i> larvae per bovine on Days -27, -25, -22, -20, -18, -15, -13, -11, -8, -4 and -1 for the purposes of randomisation and therapeutic efficacy assessments and approximately 5 000 <i>R. microplus</i> larvae on Days 21, 24, 28, 49, 52, 55, 77 and 80 for persistent efficacy assessments.	Fully engorged female ticks were collected and counted from Days 1 to 21 to assess therapeutic efficacy. Fully engorged female ticks were collected and counted from Days 42 to 56 and Days 70 to 83, to assess persistent efficacy.
388	Cattle were infested with approximately 3 000 <i>R. microplus</i> larvae per bovine on Days -27, -25, -22, -20, -18, -15, -13, -11, -8, -4 and -1 for the purposes of randomisation and therapeutic efficacy assessments and	Fully engorged female ticks were collected and counted from Days 1 to 21 to assess therapeutic efficacy, and from Days 42 to 56, Days 70 to 84 and Days 98 to 112, to assess persistent efficacy.

	approximately 5 000 <i>R. microplus</i> larvae on Days 21, 25, 28, 49, 53, 56, 77, 80 and 84 for testing persistent efficacy.	
392	Cattle were infested with approximately 30 adult unfed <i>A. variegatum</i> ticks (± 15 males and ± 15 females) on Day -6 for randomisation purposes, Day -1 for therapeutic efficacy and on Days 14, 28 and 42 to assess persistent efficacy. During tick removal, up to five male ticks were left attached to the animal. The exact number of remaining male ticks left on the animal were recorded. Prior to the subsequent tick infestation, an <i>in situ</i> count was performed to confirm the number of attached male ticks and this number was subtracted from the number of male ticks being infested on that day. Additional, <i>in situ</i> tick counts were performed at 48 h (± 4 h) and 72 h (± 4 h) after the acaricide administration or tick infestation.	Tick counts were performed at 48, 72 and 96 h (± 4 h) after infestation or treatment administration.
393	On Day -14, all goats were fitted with a corset and artificially infested with 20 (± 2) adult, unfed <i>A. variegatum</i> ticks (10 males and 10 females). Approximately 20 (± 2) adult, unfed <i>A. variegatum</i> ticks of equal sex distribution were infested for immediate therapeutic efficacy on Day -2 and for persistent efficacy on Days 14, 28, 42 and 56.	Additional <i>in situ</i> tick counts were performed at 48 (± 4 h) and 72 (± 4 h) hours after the acaricide administration or tick infestation. Tick counts and removal were performed at 96 (± 4 h) h after the acaricide administration or infestation.
390	30 (even sex distribution) adult, unfed <i>R. appendiculatus</i> ticks per ear) applied per cow on Day -6 for randomisation purposes, on Day -1 for evaluation of therapeutic efficacy, and on Days 14, 28 and 42 to assess persistent efficacy.	Ticks were removed and counted on Day -3 (72 h ± 4 h after infestation) for randomisation and ranking purposes. Ticks were counted <i>in situ</i> on Day 2 (48 h ± 4 h after acaricide treatment) and on Days 16, 30 and 44 (48 h ± 4 h after infestation) and removed on Day 3 (72 h ± 4 h after acaricide treatment) and on Days 17, 31 and 45 (72 h ± 4 h after infestation).

Supplementary Table 3. Efficacy of fipronil, fipronil and abamectin , flumethrin and fluazuron, ivermectin and closantel and cymiazole and cypermethrin against *Rhipicephalus microplus* (West and East African isolates) in the exploratory studies (nos. 385 & 387)

Day	West Africa						East Africa					
	Fipronil		Cymiazole and cypermethrin		Ivermectin and closantel		Fipronil and abamectin		Cymiazole and cypermethrin		Fluazuron and flumethrin	
	Mean	Effic.	Mean	Effic.	Mean	Effic.	Mean	Effic.	Mean	Effic.	Mean	Effic.
0 - 21	169.3	84.6	553.0	49.7	280.0	74.5	138.3	87.5	447.0	59.6	433.7	60.8
42 - 56	0.0	100	85.7	75.1	288.0	16.4	38.7	91.3	212.7	51.9	23.3	94.7
71 - 84	9.0	94.8	10.0	94.2			46.0	76.4			100.7	48.3
99 - 112	30.0	32.8	10.7	76.1								

Mean: Mean tick count; Effic.: Percent efficacy compared to untreated control

Supplementary Table 4. Efficacy of fipronil, fipronil and abamectin, against *Rhipicephalus microplus* (West and East African isolates) in the confirmatory studies (nos. 386 & 388)

Day	West Africa		East Africa	
	Fipronil		Fipronil and abamectin	
	Mean	Efficacy	Mean	Efficacy
0 to 21	4.0	91.0	21.7	93.6
42 to 56	40.0	84.3	0.0	_*
70 to 84	125.3	51.9	59.2	86.3
98 to 112	-	-	86.5	78.5

* insufficient ticks collected from the control group during this period

Mean: Mean tick count; Efficacy: Percent efficacy compared to untreated control

Supplementary Table 5. Therapeutic and persistent efficacy of fipronil and abamectin and flumethrin pour-on against *Amblyomma variegatum* in the exploratory study on goats (no.391)

Day	Timepoint	Control	Fipronil		Flumethrin pour-on	
		Mean	Mean	Efficacy	Mean	Efficacy
2	48 h post acaricide	20.0	12.7	36.7	15.3	23.3
3	72 h post acaricide	20.3	8.3	59.0	9.0	55.7
4	96 h post acaricide	19.0	7.0	63.2	0.3	98.2
16	48 h post infestation	18.3	13.0	29.1	1.0	94.5
17	72 h post infestation	18.7	10.7	42.9	1.0	94.6
18	96 h post infestation	18.3	9.0	50.9	1.0	94.5
30	48 h post infestation	19.3	16.7	13.8	3.7	81.0
31	72 h post infestation	19.3	13.7	29.3	1.7	91.4
32	96 h post infestation	19.3	13.7	29.3	1.3	93.1
44	48 h post infestation	19.3	-	-	6.9	63.8
45	72 h post infestation	19.7	-	-	7.0	64.4
46	96 h post infestation	19.7	-	-	7.0	64.4

Mean: Mean tick count; Efficacy: Percent efficacy compared to untreated control

Supplementary Table 6. Therapeutic and persistent efficacy of spray-on formulations; amitraz, cymiazole and cypermethrin, flumethrin and chlorfenvinphos and alfamethrin against *Amblyomma variegatum* in the exploratory study on goats (no.391)

Days post acaricide *	Amitraz		Cymiazole and cypermethrin		Flumethrin spray-on		Chlorfenvinphos and alfamethrin	
	Control mean (Mean)	Efficacy	Control mean (Mean)	Efficacy	Control mean (Mean)	Efficacy	Pre-count mean (post-count-mean)	Efficacy
Therapeutic efficacies								
2	20.0 (0.0)	100	18.0 (0.0)	100	19.3 (0.0)	100	19.0 (8.0)	57.9
3	20.3 (0.0)	100	18.0 (0.0)	100	19.0 (0.0)	100	19.0 (2.3)	87.7
4	19.0 (0.0)	100	18.0 (0.0)	100	19.0 (0.0)	100	19.0 (1.0)	94.7
Persistent efficacies								
10	-	-	-	-	-	-	-	-
12	-	-	19.3 (7.7)	60.3	19.3 (1.7)	91.4	-	-
13	-	-	19.3 (7.3)	62.1	19.7 (1.3)	93.2	-	-
14	-	-	19.3 (4.7)	75.9	19.7 (0.7)	96.6	-	-
16	18.3 (12.0)	34.5	-	-	-	-	-	-
17	18.7 (8.3)	55.4	-	-	-	-	-	-
18	18.3 (8.0)	56.4	-	-	-	-	-	-
24	-	-	-	-	-	-	-	-
26	-	-	-	-	19.0 (6.7)	64.9	-	-
27	-	-	-	-	19.0 (5.3)	71.9	-	-
28	-	-	-	-	19.0 (3.7)	80.7	-	-

Mean: Mean tick count; Efficacy: Percent efficacy compared to untreated control. Values are shown for the control group and the acaricide treated group in brackets.

Supplementary Table 7. Therapeutic and persistent efficacy of 1% flumethrin pour-on on goats and 2% flumethrin spray-on on cattle against *Amblyomma variegatum* in the confirmatory studies (nos. 392 & 393)

Day	Timepoint	Control		Flumethrin spray-on (cattle)		Control		Flumethrin pour-on (goats)	
		Mean	Mean	Mean	Efficacy	Mean	Mean	Mean	Efficacy
-2									Tick Challenge
2	48 h post acaricide	17.5	0.2	99.0	14.8	9.8			33.7
3	72 h post acaricide	18.0	0.0	100	14.3	3.8			73.3
4	96 h post acaricide	18.0	0.0	100	14.2	0.3			97.6
14									Tick Challenge
16	48 h post infestation	28.5	17.7	38.0	19.3	2.8			85.3
17	72 h post infestation	28.7	13.5	52.9	18.5	1.7			91.0
18	96 h post infestation	29.0	13.5	53.4	18.5	0.3			98.2
25		Acaricide administration			-	-	-		-
28									Tick Challenge
30	48 h post infestation	26.5	3.2	88.1	18.5	3.3			82.0
31	72 h post infestation	24.8	0.5	98.0	18.2	1.2			93.6
32	96 h post infestation	26.2	0.0	100	18.3	0.8			95.5
35		Acaricide administration			-	-	-		-
42									Tick Challenge
44	48 h post infestation	22.5	9.8	56.3	19.2	5.0			73.9
45	72 h post infestation	22.5	8.2	63.7	19.2	3.3			82.6
46	96 h post infestation	23.2	7.2	69.1	19.8	2.5			87.4
56									Tick Challenge
58	48 h post infestation	-	-	-	19.6	8.0			59.2
59	72 h post infestation	-	-	-	19.6	6.0			69.4
60	96 h post infestation	-	-	-	20.0	6.0			70.0

Mean: Mean tick count; Efficacy: Percent efficacy compared to untreated control

Supplementary Table 8. Therapeutic and persistent efficacy of pour-on formulations; 0.9% fipronil and 0.5% abamectin and 1% fipronil against *Rhipicephalus appendiculatus* ticks in the exploratory study (no. 389)

Day	Timepoint	Control	Fipronil and abamectin		Fipronil	
		Mean	Mean	Efficacy	Mean	Efficacy
1	24 h post acaricide	40.7	34.0	16.4	0.0	100
2	48 h post acaricide	56.3	0.3	99.4	0.0	100
16	48 h post infestation	41.0	1.0	97.6	2.0	95.1
30	48 h post infestation	36.3	1.7	95.4	9.0	75.2
58	48 h post infestation	17.0	15.0	11.8	7.7	54.9
65	24 h post infestation	34.3	18.0	47.6	18.7	45.6
66	48 h post infestation	28.7	24.7	14.0	22.3	22.1

Mean: Mean tick count; Efficacy: Percent efficacy compared to untreated control

Supplementary Table 9. Therapeutic efficacy of spray-on formulations; flumethrin dip and spray, amitraz, cymiazole and cypermethrin and chlorfenvinphos and alfamethrin against *Rhipicephalus appendiculatus* ticks in the exploratory study (no. 389)

Timepoint	Active Ingredient	Control	Acaricide	
		Mean	Mean	Efficacy
24 h post acaricide	Flumethrin	40.7	0.0	100
	Amitraz	41.0	0.0	100
	Cymiazole and cypermethrin	36.3	3.0	91.7
	Chlorfenvinphos and alfamethrin	17.0	11.3	33.3
	Amitraz	34.3	0.0	100

Mean: Mean tick count; Efficacy: Percent efficacy compared to untreated control

Supplementary Table 10. Therapeutic and persistent efficacy of fipronil and abamectin against *Rhipicephalus appendiculatus* ticks in the confirmatory study (no. 390)

Day	Timepoint	Control	Fipronil and abamectin	
		Mean	Mean	Efficacy
2	48 h post acaricide	48.3	27.8	42.4
3	72 h post acaricide	54.7	1.8	96.6
16	48 h post infestation	44.7	3.2	92.9
17	72 h post infestation	46.7	0.2	99.6
30	48 h post infestation	47.0	14.8	68.4
31	72 h post infestation	43.3	4.0	90.8
44	48 h post infestation	37.2	19.8	46.6
45	72 h post infestation	32.5	9.0	72.3

Mean: Mean tick count; Efficacy: Percent efficacy compared to untreated control