

Equine disease control Surveillance report

JD Grewar, CT Weyer & LS van Helden 2023







Surveillance Report	
Disease	Dourine in Equids
Coverage Area	Western Cape – AHS Surveillance Zone
Period	2023 – June and December
Authors	John Grewar, Camilla Weyer and Lesley van Helden

Introduction

Since 2018 active dourine surveillance has taken place in the Western Cape Province in the form of the testing of either sentinel horses (sourced from African horse sickness (AHS) sentinel program) or formal surveys of randomly selected horses in the AHS free zone (2020). The intent has been to perform surveillance in sentinel animals at 6 monthly intervals. The program in 2018 and 2019 was only performed in the first half of these years respectively. In 2020 the targeted AHS free zone survey was performed in the first half of the year as well. In 2021 and 2022 the biannual target was achieved with sentinels tested from the June and December cohorts. This has again been achieved in the same manner in 2023, the period evaluated in this report. An introduction to dourine, and the reason surveillance is required, has been thoroughly described in previous reports – available through links above or at https://www.myhorse.org.za. The scope of this report is to provide evidence for freedom of dourine within the same area where active surveillance is undertaken against AHS, i.e. the AHS surveillance and free zone in the Western Cape Province.

Surveillance parameters

Table 1: Surveillance parameters used in design and evaluation of the Dourine surveillance program in 2023.

Parameter	Value	Comments
Population at risk	16000	All horses in the surveillance and free zones. 16000 is an estimate; there are currently 18336 and 765 horses registered in the AHS surveillance and free zones respectively although data is captured only for horses entering the area.
Design Prevalence	~5%	Minimum expected prevalence in the population should dourine occur; parameter taken from the serological survey requirements of the EU for AHS sentinel surveillance given that the same population was used for the dourine sampling.
Test Sensitivity	90%	Estimate based on best scientific guess. The sensitivity of the complement fixation test (CFT) has not been established, although given the false positive rates (see specificity) the sensitivity is likely to be relatively high. The CFT is seen as a gold standard for individual horse testing prior to export and this also supports a test with relatively good sensitivity.
Test Specificity	Unknown but system specificity of 100% assumed	The CFT test is prone to false positives and probably does not have a particularly good specificity. However, for the purposes of surveillance evaluation and given that any positive CFT result will be investigated to establish a final diagnosis a specificity of 100% was used in establishing the outcome of the sensitivity of the surveillance
Type 1 error	5%	Used to provide a final probability of 95% that Dourine was not present if it was not detected using the surveillance parameters.

South Africa's sentinel surveillance program for AHS in the AHS free and surveillance zones has a target of approximately 150 horses per month using PCR testing. Serum samples are however also taken from all sentinels and a random selection from these is used for the Dourine surveillance system from the June and December sample each year. The AHS sentinel surveillance program makes every effort





to sample horses in proportion to their relative underlying population at risk using a gridded approach – see Figure 1)

Consent and Ethics

Ethics approval was not obtained nor required for this survey since the samples were used from the existing AHS sentinel surveillance program. Sentinel owners and managers had previously provided consent for this surveillance program.

Results

A total of 100 horses were selected from 36 locations across the AHS surveillance zone in June 2023. In December 100 horses were selected from 34 locations. Proportional numbers of horses sampled across the surveillance zone are shown in Figure 1 below. Most samples were taken from an area of approximately 50-75 km around the Kenilworth Quarantine Station, from which horses are exported.

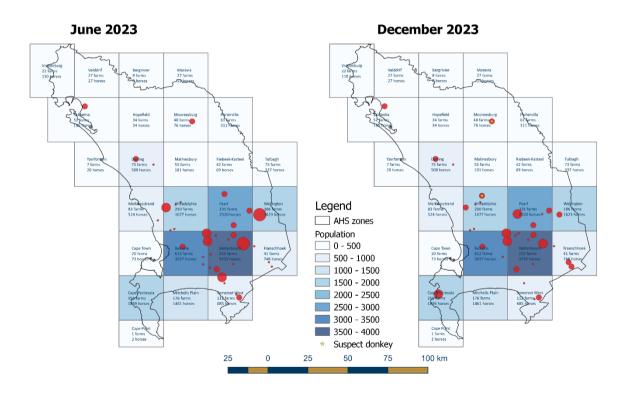


Figure 1: Dourine survey locations showing proportional circles for number of equines tested per location in June and December 2023 respectively. The underlying population at risk is shown as a light to dark blue gradient, this to show that locations were chosen to reflect the relative underlying population at risk per surveillance grid. Stars in the December panel indicate the locations of the two suspect Donkeys.

June 2023: 98 of the 100 samples tested negative for dourine antibody using the CFT (See Annex June 2023 Results), with two samples returning anti-complementary results. Both animals that returned anti-complementary results had been tested twice in 2022 within negative results.





Dec 2023: 98 of the 100 samples tested negative for dourine antibody using the CFT (see Annex December 2023 results), with two samples returning positive results. Both animals (ID's 1982 and 41938) were donkeys and information on these animals is provided below.

Animal 1982 (a ~ 7 year old jenny) had previously tested negative in this program in 2022 (both surveillance events) and in June 2023. On investigation and consultation with the farm manager the following was established

- The animal has been on the property for 6-7 years and has not had contact with stallions nor jacks. It was originally sourced from Piketberg.
- It has not shown clinical signs consistent with Dourine.
- Two other animals on the farm are sentinels and tested Dourine negative in the Dec 2023 surveillance round.

Animal 41938 (a ~20 year old castrated jack) was a new sentinel and had not previously been included in a Dourine surveillance event. On investigation and consultation with the farm manager the following was established:

- The animal, in very poor condition at the time) was rescued from a farm in Malmesbury in mid 2023.
- It stands with 2 ponies and a horse and does not have sexual contact with these animals.
- In the last five months no overt signs of Dourine have been noted.
- The three other animals on the farm are sentinels and tested Dourine negative in the Dec 2023 surveillance round.

Both animals were sampled on 28 March 2024 during follow up investigations. Both samples in March returned Dourine suspect results (see Annex March 2024 Results – Follow up).

On further evaluation of the <u>WOAH manual for Dourine</u> it is clear that donkeys and mules may remain inapparent carriers, but also that these species often give inconsistent or nonspecific reactions because of the anticomplementary effects of their sera. Preliminary conclusions are that animal 1982 is highly likely to be a false positive given its history and contact situation. Animal 41938 is unlikely to be a true case given its origin in the AHS surveillance zone, however the lack of previous testing and recent introduction on to the farm cannot rule out Dourine explicitly. The animal is however castrated and poses no threat to other animals in relation to Dourine and this animal will be included in the June and December 2024 submission to try get a more extensive testing profile.

Sensitivity of the surveillance program

The sensitivity (and resulting probability of freedom) of the surveillance program is shown in Table 2 below. This evaluation is independent of any prior surveillance. While the sentinel surveillance program is based on a single stage sampling strategy (evaluated in Column 2 of Table 2) we have estimates of the underlying number of herds in the surveillance zone as well as estimates of the herd sizes of the sampled herds. This allows an estimate of surveillance sensitivity in a more realistic setting (Column 3 of Table 2). Note that in this latter analysis we reverted to an effective population design





prevalence of 2% (within herd design prevalence of 20% and herd level prevalence of 10% throughout the population) – this in an effort to depict a reasonable minimum expected prevalence with so few cases of dourine reported in the prior two decades in the AHS surveillance zone (see Figure 3).

Table 2: Design prevalence with resulting surveillance sensitivity and probability of freedom outcomes for two different scenarios independently analysed: the sentinel program design prevalence and the generic values used given the history of cases in the AHS controlled area. NOTE: This evaluation is for a single point in time and does not consider previous surveillance outcomes.

Parameter	Descriptions and values based on varying data sources			
	Single stage population	n sensitivity	1	to result in effective % with 2-stage analysis
Animal level prevalence (P*u)	0.05	5	().2
Herd level prevalence (P*c)	n/a		C).1
Effective population prevalence (P*u x P*c)	0.05	5	0	.02
	June 2023	December 2023	June 2023	December 2023
<i>MeanSSH</i> - Mean herd level surveillance sensitivity	n/a	I	0.439	0.476
SeP - Population surveillance sensitivity	0.988	0.988	0.797	0.805
PFreeU - Confidence of population freedom – uninformed prior	0.985	0.985	0.801	0.807

With surveillance evaluation it is also appropriate to evaluate probability of freedom outcomes given prior surveillance events. In Figure 2 below, this evaluation shows all 9 surveillance events undertaken to date in the AHS surveillance and free zones (see previous reports). Where surveillance was missed (second half of 2018 (H2), 2019 (H4) and 2020 (H6)) a zero sensitivity is assumed. Note also that the surveillance in 2020 (H5) was targeting the AHS free zone only, but the evaluation below assumes a population at risk across the AHS free and surveillance zone for standardization purposes.

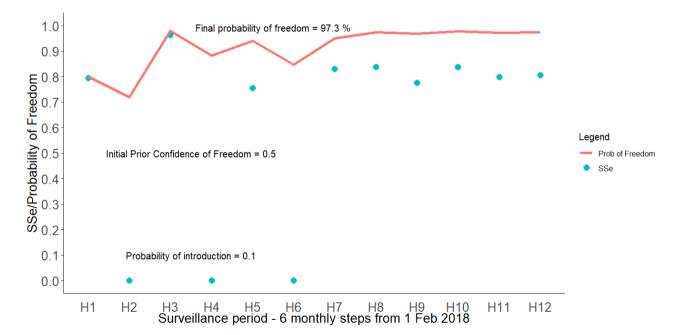


Figure 2: Surveillance system sensitivity and probability of freedom assuming an introduction probability of 10%, an underlying herd and animal prevalence of 20 % and 10% respectively and an uninformed prior probability of 50% in period 1.





The dourine probability of freedom in the AHS free and surveillance zone in June and December 2023, given the 2023 surveillance efforts alone, ranges between 80% and 98.5% depending on the analysis used. The overall probability of freedom taking prior surveillance into account is 97.3%.

Discussion

Stand-alone surveillance efforts like the one described here supplement the current clinical passive surveillance and Thoroughbred pre-breeding dourine surveillance efforts in South Africa.

Figure 3 shows all dourine cases reported in South Africa from 1993 through Jan 2018 (data accessed Apr 2024 from <u>www.dalrrd.gov.za</u> and collated to South African local municipalities). The last case in the Western Cape in 2012 occurred in a working mule in the Bredasdorp region – details of that case can be found at <u>http://www.elsenburg.com/vetepi/epireport_pdf/February2012.pdf</u>.

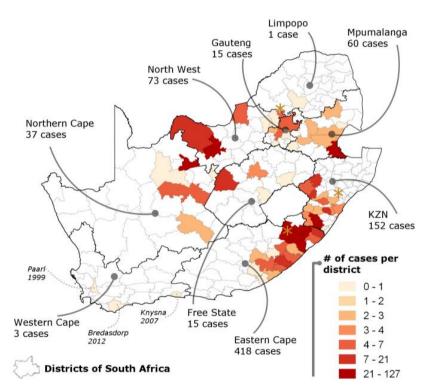


Figure 3: Historical dourine cases reported to DALRRD from 1993 through Jan 2018 (the last case as reported by DALRRD) to date. Cases have been aggregated by district while case totals per province are labelled. The three cases reported in the Western Cape are also labelled specifically with the <u>last case reported in 2012</u>.

Acknowledgments

We are very grateful to our colleagues in the Department of Agriculture, Land Reform and Rural Development, (DALRRD), the Western Cape Department of Agriculture and the ARC - Onderstepoort Veterinary Research who facilitated this work. Funding for this project was obtained from the South African Equine Health and Protocols (SAEHP - sampling, logistic and testing costs) and the Western Cape Department of Agriculture (sample kits). We are, as always, very grateful to the owners and managers of the sentinel horses in the AHS controlled area in the Western Cape.





June 2023 Results



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BACTERIAL SEROLOGY

2023-D-13014 30/08/2023 31/08 A 11/09/2023 Serum

IJac MR

31/08/2023 to 07/09/2023 100 SURVEILLANCE SUITABLE

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FINAL REPORT: Dourine Complement Fixation Test (CFT) Method BSME005

Sample No.	Species	Sample Reference	Results
1	Equine	Canta Libre 8564	Negative
2	Equine	Kentucky 1685	Negative
3	Equine	Saartjie 1982	Negative
4	Equine	Bono 35342	Negative
5	Equine	Calif Stonewalls Choice 30831	Negative
6	Equine	Rango 14832	Negative
9	Equine	Chantilly 1687	Negative
10	Equine	Priya Pazice 26562	Negative
11	Equine	Ransheen 25495	Negative

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Sample No.	Species	Sample Reference	Results
12	Equine	Sultan (Muter) 1689	Negative
15	Equine	Beauty 5806	Negative
16	Equine	Shannon 1529	Negative
17	Equine	Leyhla Kamana 28883	Negative
18	Equine	Leyhla Krimson (17 Jewel) 19616	Negative
7	Equine	Victor 1629	Negative
8	Equine	Alfadee 25494	Negative
21	Equine	Jerico Allegra 23470	Negative
22	Equine	Daisy 22548	Negative
23	Equine	Poppie 18171	Negative
24	Equine	Valentino 28730	Negative
13	Equine	Warren Hill (Lazuli) 1690	Negative
14	Equine	Sultan 1504	Negative
27	Equine	Tsalta Echo 12934	Negative
28	Equine	Geyerspan Haraldi 14943	Negative
29	Equine	Jurust 66 Dakar 15352	Negative
30	Equine	Twister 14944	Negative
19	Equine	Leyhla Krystal (17 Fantasy) 19617	Negative
20	Equine	Leyhla Thunder Storm (17 Kyte) 19615	Negative
33	Equine	Daantjie 6091	Negative
34	Equine	Gazalle 6468	Negative
35	Equine	Legacys La Sagra : Cassandra 34293	Negative
36	Equine	Legacys Lucky Dragon 6072	Negative
25	Equine	Moniqi Zair 20780	Negative
26	Equine	Ruah Zaur 35689	Negative
39	Equine	Lily 1511	Negative
40	Equine	Brandy 33041	Negative
41	Equine	Eagles Colour Code 15827	Negative
42	Equine	Lily 35690	Negative
31	Equine	Bomalia 6077	Negative

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Sample No.	Species	Sample Reference	Results
32	Equine	Calypso 6073	Negative
45	Equine	Amado 25863	Negative
46	Equine	Big Mamma (Madison) 25862	Negative
47	Equine	Charlesto 25866	Negative
48	Equine	Hot Shot (Santiago) 25861	Negative
37	Equine	Luna 23226	Negative
38	Equine	Real Rose 6096	Negative
51	Equine	Winnipeg 33794	Negative
52	Equine	Areshak Rugaya 9685	Negative
53	Equine	Shilo 34007	Negative
54	Equine	The Sequel 39992	Negative
43	Equine	Vrystaat 39979	Negative
44	Equine	Spirit 1510	Negative
57	Equine	Rivendale Count Felix 1774	Negative
58	Equine	Sonic 8611	Negative
59	Equine	Billy Bob 31770	Negative
60	Equine	Midnight Sky 38476	Negative
49	Equine	Peppadew (Dixie) 25865	Anti-complement
50	Equine	Ruah Baruk 25864	Negative
63	Equine	Grey (Never Imagined) 1535	Negative
64	Equine	Leomardo 30592	Negative
65	Equine	Pink Lady 36931	Negative
66	Equine	Ariel 5937	Negative
55	Equine	The Higgs 30192	Negative
56	Equine	Wizard 472	Negative
69	Equine	Southcross Beowulf 29147	Negative
70	Equine	Waterford Compose (Libby) 8918	Negative
71	Equine	Kelty 1672	Negative
72	Equine	Bahlu 36873	Negative
61	Equine	Milo 23465	Negative

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Sample No.	Species	Sample Reference	Results
62	Equine	Borrasca 30617	Negative
75	Equine	Res 36871	Negative
76	Equine	Suki 36870	Negative
77	Equine	Davetsaub Alex 5783	Negative
78	Equine	Elsa 26564	Negative
67	Equine	Blue 35282	Negative
68	Equine	Gummy 35283	Negative
81	Equine	Numero uno Zakoen (Sabek) 5782	Negative
82	Equine	Parfait Finesse 26561	Negative
83	Equine	Ice Man 24273	Negative
84	Equine	Talula 24272	Negative
73	Equine	Fly Baby Fly 16286	Negative
74	Equine	Nyala 368872	Negative
87	Equine	RK Crysos 35417	Negative
88	Equine	RK Eliseo 32214	Negative
89	Equine	RK Sorpresa 35777	Negative
90	Equine	RK Venteux 35778	Negative
79	Equine	Fine Pearl 5831	Negative
80	Equine	Fleur (Blommie) 16648	Negative
93	Equine	Klippies 1635	Negative
94	Equine	Emmy (Making Magic) 1791	Negative
95	Equine	Mr Goss (Big) 23143	Negative
96	Equine	Hercules 1655	Negative
85	Equine	Lumiere Lis 27861	Negative
86	Equine	Othello 35416	Negative
97	Equine	Little Bits 1681	Anti-complemen
98	Equine	Almarada Leila 12633	Negative
99	Equine	Caesar PW 18958	Negative
100	Equine	Silver Honey Bear 18963	Negative
91	Equine	Dawie 5830	Negative

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92 Equine	Charlotte 1633	Negative
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Comment: It is recommended that the infection status of animals with test results other than negative be confirmed by repeat testing.

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Your feedback on our service is valuable. Please contact us with any recommendations for improvement of the anomalies on the report.

Samples(s) Submitted was/were suitable for testing Yours Faithfully

Photour

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Maria Mathekga Technical Signatory for Senior Manager Research: Animal Health and Protection

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December 2023 results



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Job No: Received Date: Lab No: Report Date: Sample Type: Sample collection date: Performance Date: Number of samples: Purpose of Sampling: Suitability of samples:

> **Owner Details** Owner: Email:

Farm:

Various

BACTERIAL SEROLOGY

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> > 2024-D-02102 19/02/2024 19/02 F 08/03/2024 Serum 22/02/2024 to 29/02/2024

Hac-MRA

100 SURVEILLANCE SUITABLE

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FINAL REPORT: Dourine Complement Fixation Test (CFT) Method BSME005

Sample No.	Species	Sample Reference	Results
1	Equine	CANTA LIBRE 8564	Negative
2	Equine	KENTUCKY 1685	Negative
3	Equine	SAARTJIE 1982	Positive
4	Equine	BONO 35342	Negative
5	Equine	CALIF STONEWALSS CHOICE 30831	Negative
6	Equine	RANGO 14832	Negative
9	Equine	CHANTILLY 1687	Negative
10	Equine	RANSHEEN 25495	Negative
11	Equine	SULTAN (MUTER) 1689	Negative
12	Equine	LILY 1511	Negative

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Sample No.	Species	Sample Reference	Results
15	Equine	LEYHLA KRYSTAL (17 FANTASY) 19617	Negative
16	Equine	LEYHLA THUNDER STORM (17KYTE) 19615	Negative
17	Equine	BRANDY 33041	Negative
18	Equine	EAGLES COLOUR CODE 15827	Negative
7	Equine	VICTOR 16929	Negative
8	Equine	ALFADEE 25494	Negative
21	Equine	DAISY 22548	Negative
22	Equine	POPPIE 18171	Negative
23	Equine	VALENTINO 28730	Negative
24	Equine	SPIRIT 1510	Negative
13	Equine	BEAUTY 5806	Negative
14	Equine	LEYHLA KAMANA 28883	Negative
27	Equine	PONY 24274	Negative
28	Equine	TALULA 24272	Negative
29	Equine	AMADO 25863	Negative
30	Equine	BIG MAMMA (MADISON) 25862	Negative
19	Equine	LILY 35690	Negative
20	Equine	VRYSTAAT 39979	Negative
33	Equine	RUAH BARUK 25864	Negative
34	Equine	WINNIPEG 33794	Negative
35	Equine	ARESHAK RUGAYA 9685	Negative
36	Equine	SATI 1524	Negative
25	Equine	DONKEY 41938	Positive
26	Equine	ICE MAN 24273	Negative
39	Equine	MAYA 41994	Negative
40	Equine	SHILO 34007	Negative
41	Equine	THE HIGGS 30192	Negative
42	Equine	WIZARD 472	Negative
31	Equine	CHARLESTON 25866	Negative
32	Equine	PEPPADEW (DIXIE) 25865	Negative

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Sample No.	Species	Sample Reference	Results
45	Equine	SONIC 8611	Negative
46	Equine	WILLOW (LADY WILOWAY) 1568	Negative
47	Equine	CANDY 26565	Negative
48	Equine	CHAMPAGNE 40	Negative
37	Equine	NAKHIRI 1522	Negative
38	Equine	ISY 41993	Negative
51	Equine	DAANTJIE 6091	Negative
52	Equine	LAGACYS LUCKY DRAGON 6072	Negative
53	Equine	GEYERSPAN HARALDI 14943	Negative
54	Equine	JURUST 66 DAKAR 15352	Negative
43	Equine	LALI 14953	Negative
44	Equine	RIVENDALE COUNT FELIX 1774	Negative
57	Equine	KIRA 41377	Negative
58	Equine	MIDNIGHT SKY 38476	Negative
59	Equine	MILO 23465	Negative
60	Equine	BORRASCA 30617	Negative
49	Equine	BOMALIA 6077	Negative
50	Equine	CALYPSO 6073	Negative
63	Equine	ARIEL 5937	Negative
64	Equine	BLUE 35282	Negative
65	Equine	GUMMY 35283	Negative
66	Equine	SOUTHCROSS BEOWUKF 29147	Negative
55	Equine	TWISTER 14944	Negative
56	Equine	AYLA 38940	Negative
69	Equine	ALADDIN 41992	Negative
70	Equine	BEN 41147	Negative
71	Equine	EAST COAST 41144	Negative
72	Equine	LIEFIE 41146	Negative
61	Equine	GREY (NEVER IMAGINED) 1535	Negative
62	Equine	ISABELLE 36612	Negative

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Sample No.	Species	Sample Reference	Results
75	Equine	DAVETSAUB ALEX 5783	Negative
76	Equine	FINE PEARL 5831	Negative
77	Equine	FLEUR (BLOMMIE) 16648	Negative
78	Equine	NUMERO UNO ZAKOEN (SABEK) 5782	Negative
67	Equine	WATERFORD COMPOSE (LIBBY) 8918	Negative
68	Equine	RAZ 1514	Negative
81	Equine	LUNIERE LIS 27861	Negative
82	Equine	OTHELLO 35416	Negative
83	Equine	RK CRYSOS 35417	Negative
84	Equine	RK ELISEO 32214	Negative
73	Equine	SPROET 41146	Negative
74	Equine	STARLIGHT 28836	Negative
87	Equine	CHARLOTTE 1633	Negative
88	Equine	KLIPPIES 1635	Negative
89	Equine	CAESAR PW 18958	Negative
90	Equine	MOSCOW MAGIC 15586	Negative
79	Equine	PARFAIT FINESSE 26561	Negative
80	Equine	JASMYN 34708	Negative
93	Equine	AVATAR 36932	Negative
94	Equine	BENTLEY 36933	Negative
95	Equine	GHOST 31270	Negative
96	Equine	JUPITERS JUNO 40989	Negative
85	Equine	RK SORPRESA 35777	Negative
86	Equine	RK VENTEUX 35778	Negative
97	Equine	RAKA 31272	Negative
98	Equine	RUBY MOON 40990	Negative
99	Equine	CEO (ALI) 35779	Negative
100	Equine	HUNTERS MOON 35781	Negative
91	Equine	ZANXIBAR 33791	Negative
92	Equine	LITTLE BITS 1681	Negative

2024-D-02102

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Comment: It is recommended that the infection status of animals with test results other than negative be confirmed by repeat testing.

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Your faedback on our service is valuable. Please contact us with any recommendations for improvement of the anomalies on the report.

Samples(s) Submitted was/were suitable for testing Yours Faithfully

Maria Mathekga

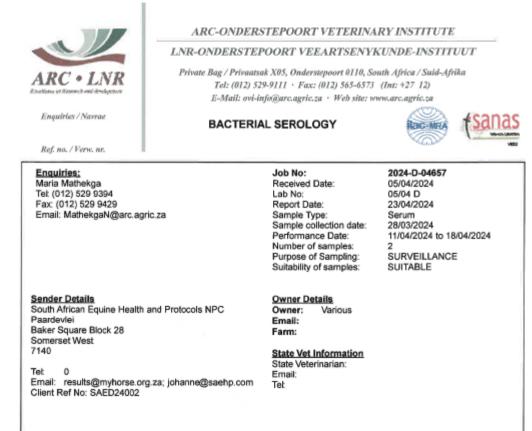
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March 2024 Results - Follow up



FINAL REPORT: Dourine Complement Fixation Test (CFT) Method BSME005

Sample No.	Species	Sample Reference	Results
1	Equine	DONKEY - LSS 240 037	Suspect
2	Equine	SAARTJE - LSS 240 038	Suspect

Comment: It is recommended that the infection status of animals with test results other than negative be confirmed by repeat testing.

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