

QC SEROLOGY INVESTIGATION

PMM-QC-885

NRL is a:

- **Accredited for compliance with ISO 17043, with NATA Accreditation Number 14253;**
- **A World Health Organization (WHO) Collaborating Centre for Diagnostics and Laboratory Support for HIV and AIDS and Other Blood-borne Infections.**

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1 INTRODUCTION

During daily out of range (OOR) and monthly data reviews of EDCNet, a number of ‘random drops’ were noted for the Optitrol HIV Screening QC testing on the Grifols PANTHER Ultrio Elite HBV/HCV/HIV assay.

These events were similar to a previously identified issue of ‘random drops’ for data on the Grifols PANTHER Ultrio Elite HBV/HCV/HIV Screening assay when testing with Optitrol NAT HIV Screening QC lots DM20017 and DM20232. This investigation resulted in a review and modification of dilution factors for the Optitrol HIV Screening QC, as well as a Medical Device Recall (TGA Recall RC-2021-RN-01328-1).

It was observed that the recent random drops were still occurring across multiple new QC lots, although at a lesser frequency. There was also a request by one participant to investigate random drops on the same assay when testing with another QC product from a different manufacturer – AcroMetrix HIVRNA+ (Thermofisher Scientific). This report will look at findings for both QC products.

2 FINDINGS

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3 DISCUSSION

Data from all participants in the peer group (Grifols PANTHER Ultrio Elite HBV/HCV/HIV assay when testing the Optitrol HIV Screening QC) were investigated to establish whether there was any evidence of trends or patterns across the peer group, including for evidence to explain the ‘random drops’ in data. This investigation followed a path set by a previously identified issue with Optitrol HIV Screening QC when tested on the Grifols PANTHER Ultrio Elite HBV/HCV/HIV assay, where QC lots DM20017 and DM20232 were also displaying ‘random drops’ in data. It was concluded in the previous investigation that the root cause was a lower

viral load than expected within specific (early) production lots. The manufacturing methodology was modified to incorporate more robust methods to quantify viral load for what is essentially a QC for a qualitative assay that has a much lower limit of detection than most viral load assays.

The more recent QC production lots, DM21082, DM21088, DM21108 and DM21128 were manufactured with a 0.1% higher HIV viral load concentration (previous lots were manufactured with a HIV viral load concentration of 0.0396 %). The very nature of lower viral load would likely result in regular and consistent viral load levels below expectation, yet evidence suggested that in the earlier production lots, regardless of the viral load being low, there were regular 'normal' S/Co ratios, with a high rate of 'drops' in S/Co ratio. The explanation that low viral load doesn't really explain this phenomenon, but regardless, the viral load concentration was corrected anyhow. Now that viral load is at the required level, 'drops' continue to occur. In addition, a product from another QC manufacturer is also displaying similar S/Co ratio drops, indicating something else may be at the root cause of the phenomenon.

Figures 1 and 2 show peer group data for the Grifols Procleix PANTHER Ultrio Elite assay testing with Optitrol NAT HIV Screening DM21082. All four participants recorded data below QConnect Limits (8.70 – 14.30 S/Co) across three reagent lot numbers.

Figures 3 and 4 show peer group data for the Grifols Procleix PANTHER Ultrio Elite assay testing with Optitrol NAT HIV Screening DM21088. Nine participants recorded data below QConnect limits (8.70 – 14.30 S/Co) across 8 reagent lot numbers.

Figures 5 and 6 show peer group data for the Grifols Procleix PANTHER Ultrio Elite assay testing with Optitrol NAT HIV Screening DM21108. Six participants recorded data below QConnect limits (8.70 – 14.30 S/Co) across six reagent lot numbers.

Peer group data for the previously identified issue with Optitrol HIV Screening QC on the Grifols PANTHER Ultrio Elite HBV/HCV/HIV assay, where QC lots DM20017 and DM20232 were giving 'random drops' in data, shown in Figures 7 – 10.

Although all five Optitrol HIV Screening QC lot numbers show 'random drops' in data, the most recent QC lot numbers under investigation in this report did not demonstrate 'drops' as frequently as the previously known issue of a low viral load for DM20017 and DM20232. Table 1 shows the 'drop' percentage for each QC lot number, where 'drops' were identified as results that fall below the lower QConnect Limit of 8.7 S/Co. QC lots DM21082, DM21088 and DM21108 show percentage 'drops' much lower than for DM20017 and DM20232, with drops of 1.15%, 1.19%, 0.89%, 3.85% and 17.06% respectively. As viral load has been corrected, the low viral load root cause was unlikely as the root cause for the recently observed drops in the QC lot numbers since. The Optitrol HIV Screening QC lot numbers DM20017 and DM20232 will no longer be referred to for the remainder of this investigation.

NRL was informed by two participants – Participant 1 and Participant 5 – that they are also observing these 'random drops' when testing with the AcroMetrix HIVRNA+ EQC. Figures 12 and 13 show the peer group data for Grifols Procleix PANTHER Ultrio Elite when testing with AcroMetrix HIVRNA+ 221515. Random drops can be seen for both laboratories over two reagent lot numbers (702892 and 703038). The percentage of results under the QConnect Limit of 8.7 S/Co is 3.20% (Table 2).

Participants with results below the lower QConnect limit of 8.7 S/Co were investigated individually to establish whether there was any evidence of trends or patterns for the 'random drops' observed (Figures 22 – 35).

Participant 5 had a known instrument issue between June and October 2021 over two EQC lot numbers (lots DM21082 and DM21088) and several reagent lots. Occasional 'random drops' below QConnect limits for DM21088 (Figure 24) were observed, four of which were unrelated to the known instrument issue. This known instrument issue also affected the Grifols Procleix PANTHER Ultrio Elite HIV discriminatory assay (Figure 35). All other 'drops' observed, including for the AcroMetrix HIVRNA+ QC (Figure 25), appear random in nature.

Participant 378 was routinely reporting data lower than the peer group for EQC lot number DM21088 (Figure 3). Due to this, some of the results fall below the QConnect lower limit of 8.7 S/Co and thus was considered unrelated to the issue under investigation (Figure 27).

Greater variation between instruments was observed for Participant 11 (Figure 29). Although a greater number of drops was observed for this participant, due to the higher number of data points, the percentage of 'drops' overall was only 1.39% (Table 3).

Participant 446 shows a 'drop' percentage of 3.64% for EQC lot number DM21088 (Figure 32). No potential root cause was observed.

Figures 16 – 21 show peer group data for Optitrol HIV Screening QC when tested on the Grifols PANTHER Ultrio Elite HIV discriminatory assay. Only a few participants report data in EDCNet for the discriminatory assays. Random drops can also be observed infrequently Optitrol HIV Screening QC lot numbers DM21082 and DM21088. Only six participants have provided data for this assay. Excluding the known instrument issue previously mentioned for Participant 5, a percentage drop of 1.44% is observed for Optitrol HIV Screening QC DM21082 and DM21088 and AcroMetrix HIVRNA+ 221515, combined, for results below 16.0 S/Co.

The Grifols Procleix PANTHER Ultrio Elite instructions for use (IFU) indicates that several of the reagents have a high risk of forming precipitates when stored at incorrect temperatures when the temperature falls between 2 – 15C (see IFU, Reagent Preparation – page 12). Perhaps this may be something that should be discussed with Grifols.

Of the 13 reagent lots in use across the peer group (Figure 11), four started with '702', six started with '703' and three started with '704'. Only the 702xxx and 703xxx reagent lots demonstrated 'data drops' while none have yet been identified in the '704xxx' reagent lots.

Of the 20 labs that reported data in EDCNet for the Grifols PANTHER Ultrio Elite assay, 16 of them reported results below the QConnect Limit of 8.7 S/Co for HIV. As Grifols PANTHER Ultrio Elite assay is a qualitative assay, and all the random low results were above the cut-off (S/Co of 1.0), the results obtained are to be determined as reactive and should be reported as HIV RNA detected. There was no significant variation observed between the three EQC lot number DM21082, DM21088 and DM21108. All lots have similar S/Co percentage rates below 8.7.

Another QC product, Optitrol NAT Triplex – designed for the Roche cobas 6800 molecular donor screening platform – was manufactured in the same way as the Optitrol NAT HIV Screening QC yet also does not observe any variation in results as seen with Ultrio Elite, whether before or after the viral load correction (Optitrol NAT Triplex viral load was also corrected in the same way as the Optitrol NAT HIV Screening QC).

4 RECOMMENDATIONS

NRL recommends discussion with DiaMex to, once again, review the production and manufacturing procedures for Optitrol NAT HIV Screening, with the suggestion to manufacture a lot that has 0.2% HIV viral load concentration (or 500 IU/mL or ~20x limit of detection) to completely exclude once and for all viral load as the subject of scrutiny (root cause). If 'drops' continue then viral load could be reduced back to 10x limit of detection, more than sufficient for a qualitative assay QC.

NRL will also contact Grifols US in order to discuss potential solutions, including data on reagent lot manufacturing information and possible changes to components if possible.

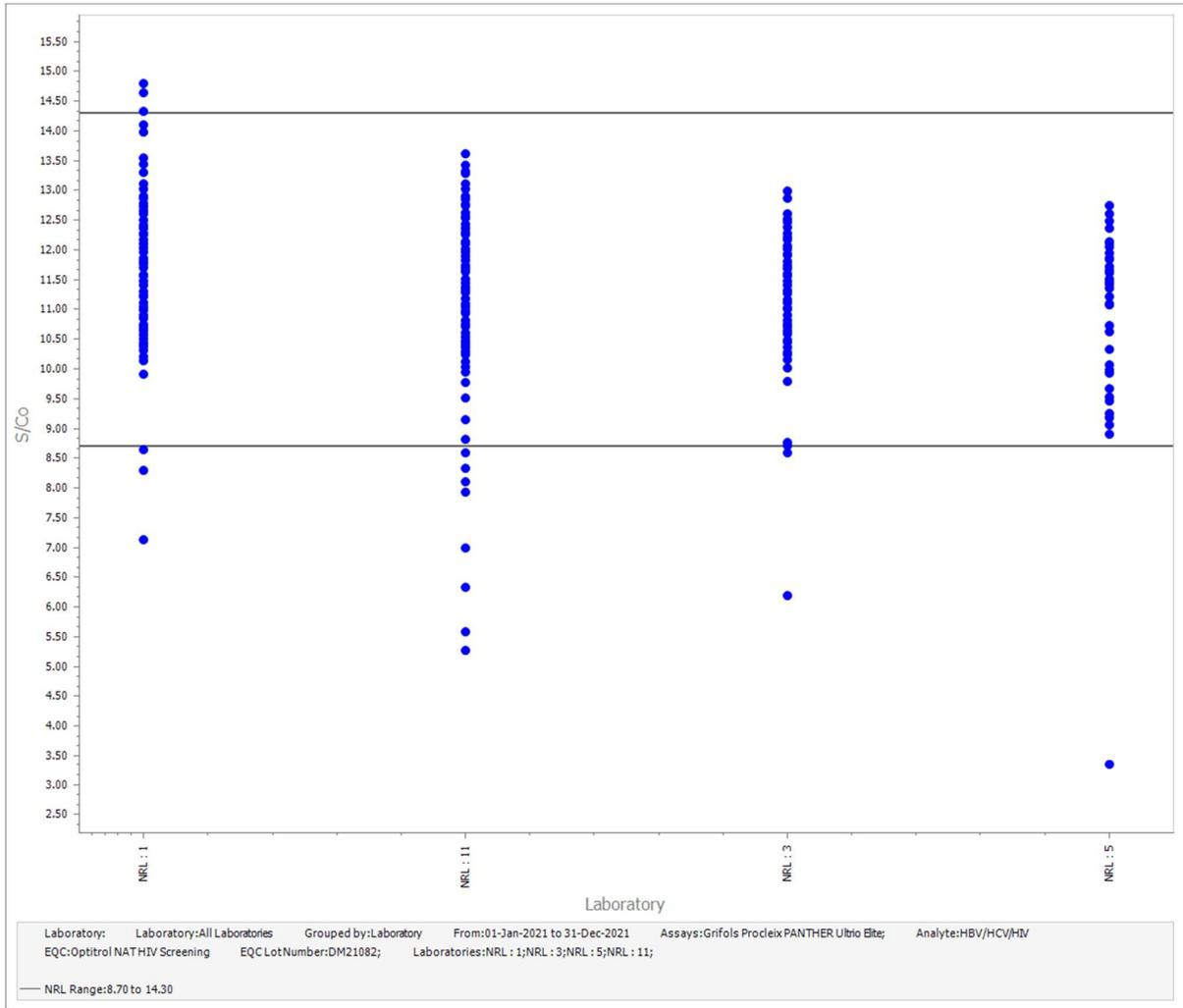


Figure 1 – Peer group data for Grifols Procleix PANTHER Ultrio Elite, Optitrol NAT HIV Screening DM21082 by participant.

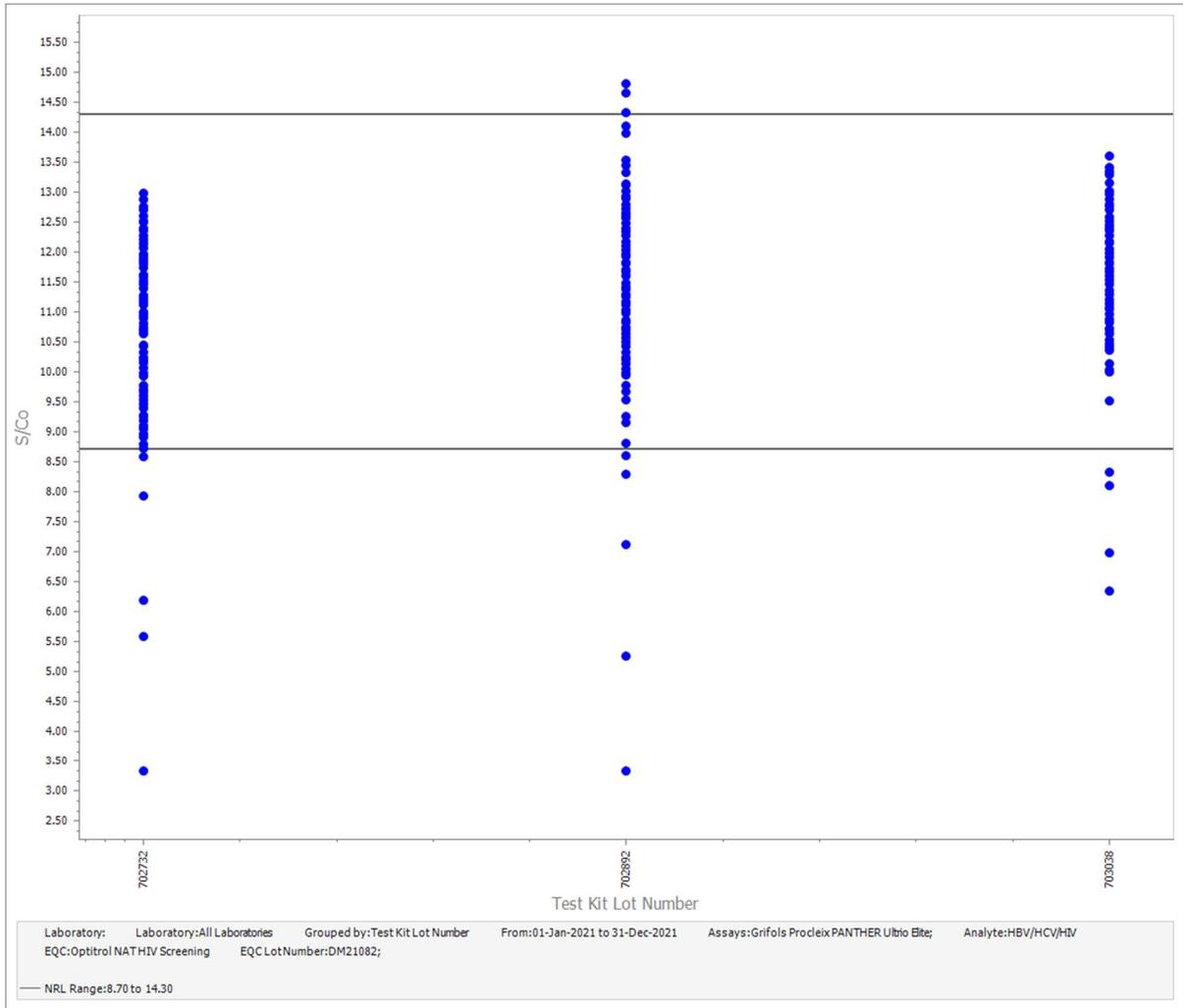


Figure 2 – Peer group data for Grifols Procleix PANTHER Ultrio Elite, Optitrol NAT HIV Screening DM21082 by reagent lot.

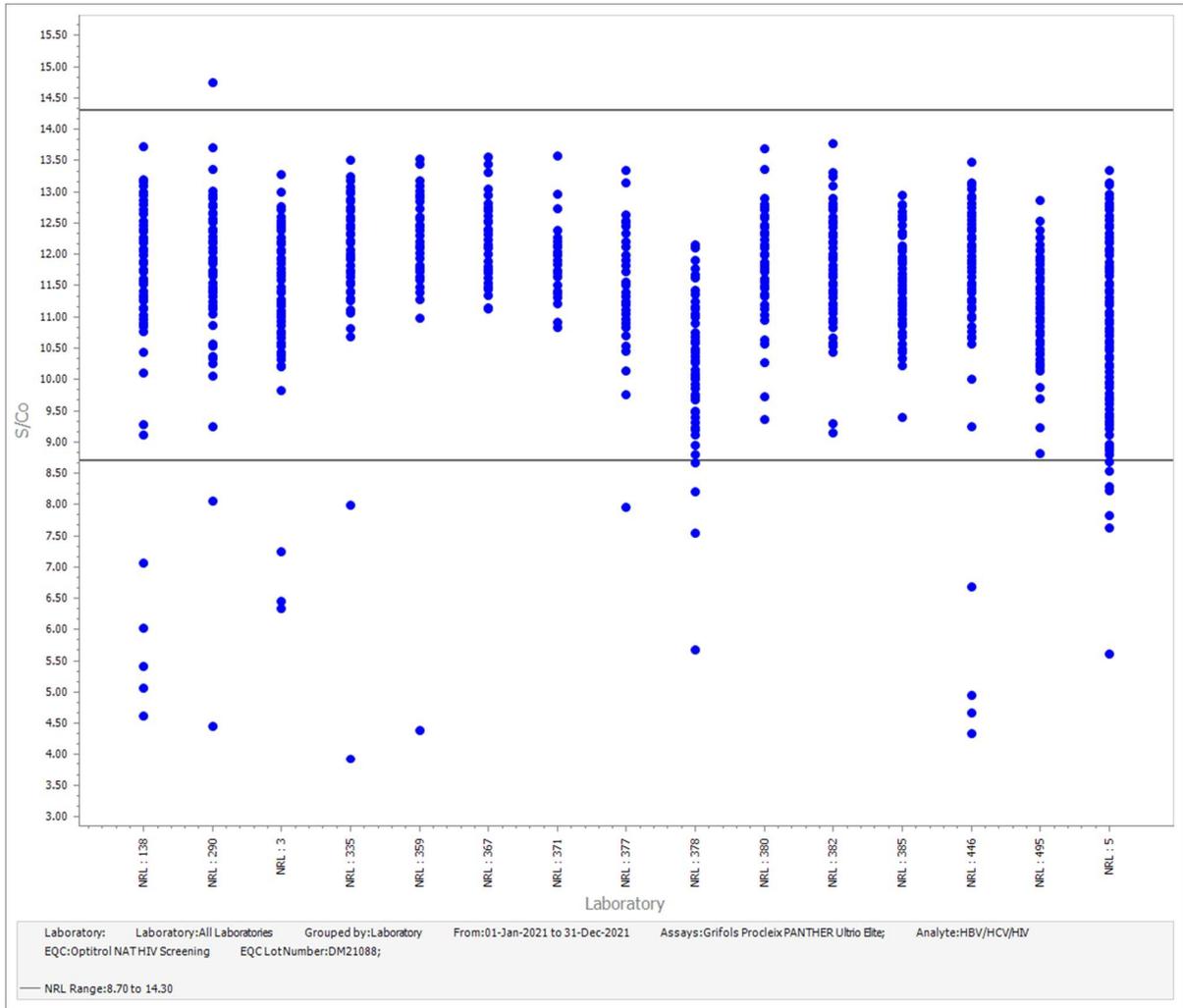


Figure 3 – Peer group data for Grifols Procleix PANTHER Ultrio Elite, Optitrol NAT HIV Screening DM21088 by participant.

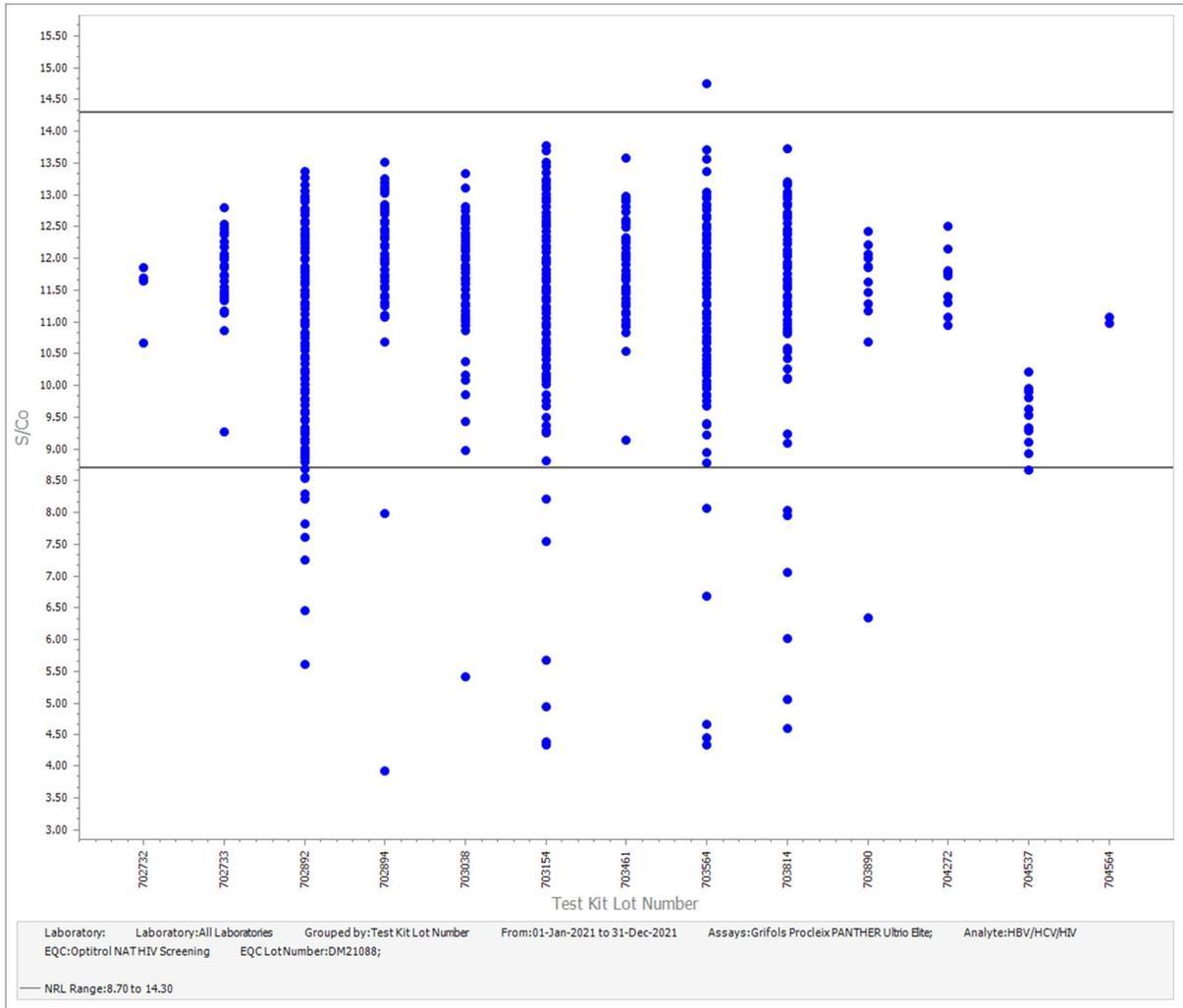


Figure 4 – Peer group data for Grifols Procleix PANTHER Ultrio Elite, Optitrol NAT HIV Screening DM21088 by reagent lot.

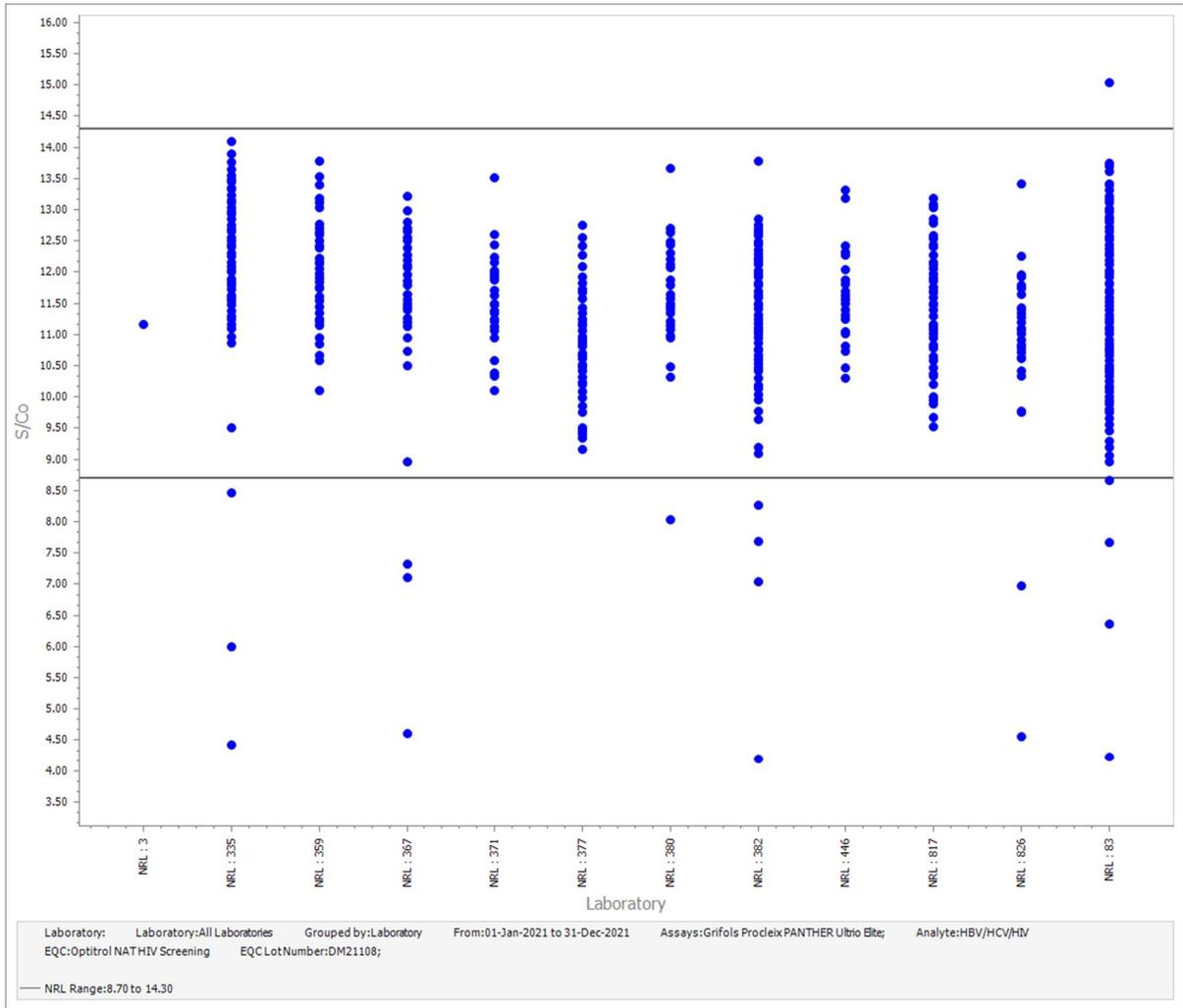


Figure 5 – Peer group data for Grifols Procleix PANTHER Ultrio Elite, Optitrol NAT HIV Screening DM21108 by participant.

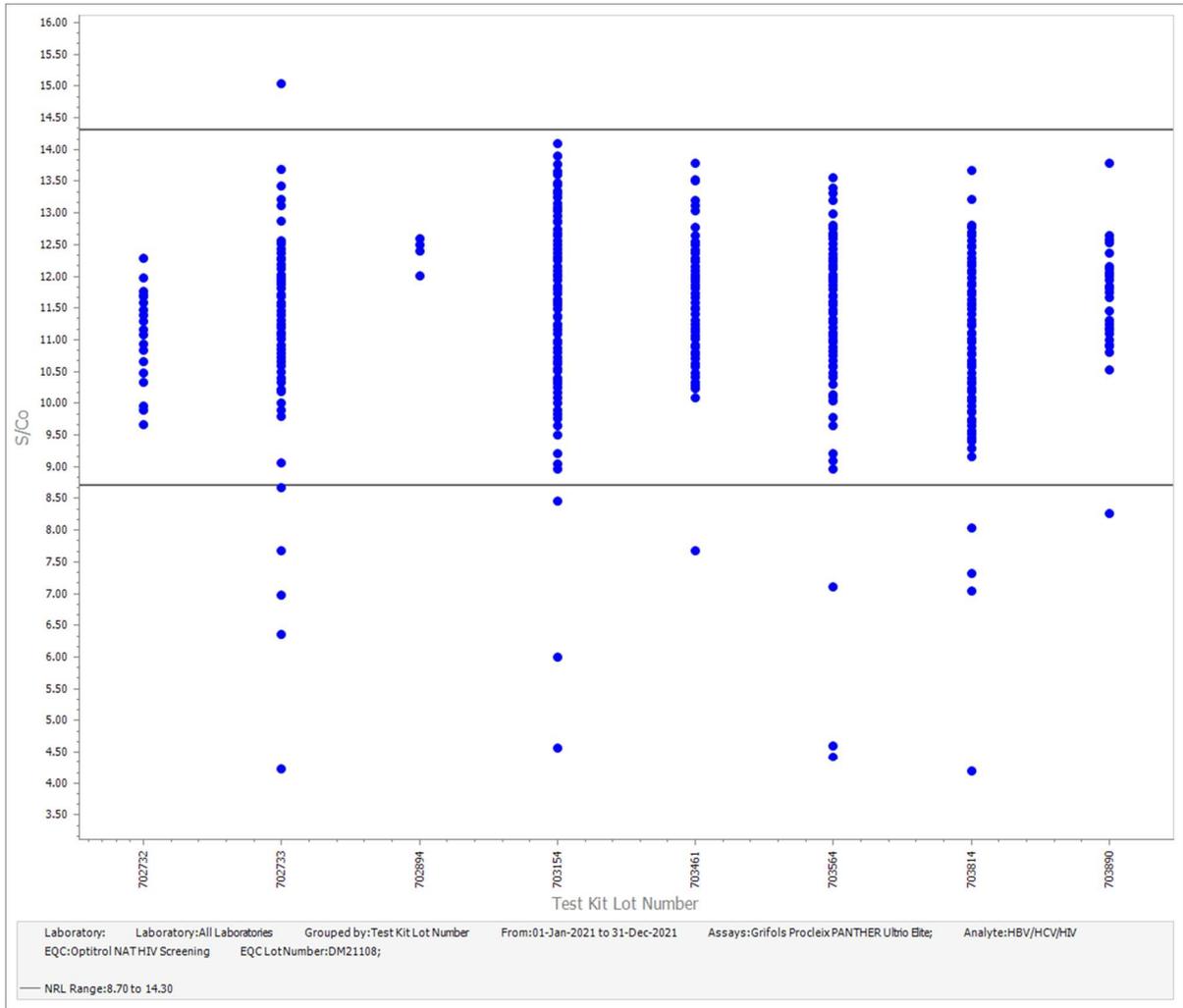


Figure 6 – Peer group data for Grifols Procleix PANTHER Ulbrio Elite, Optitrol NAT HIV Screening DM21108 by reagent lot.

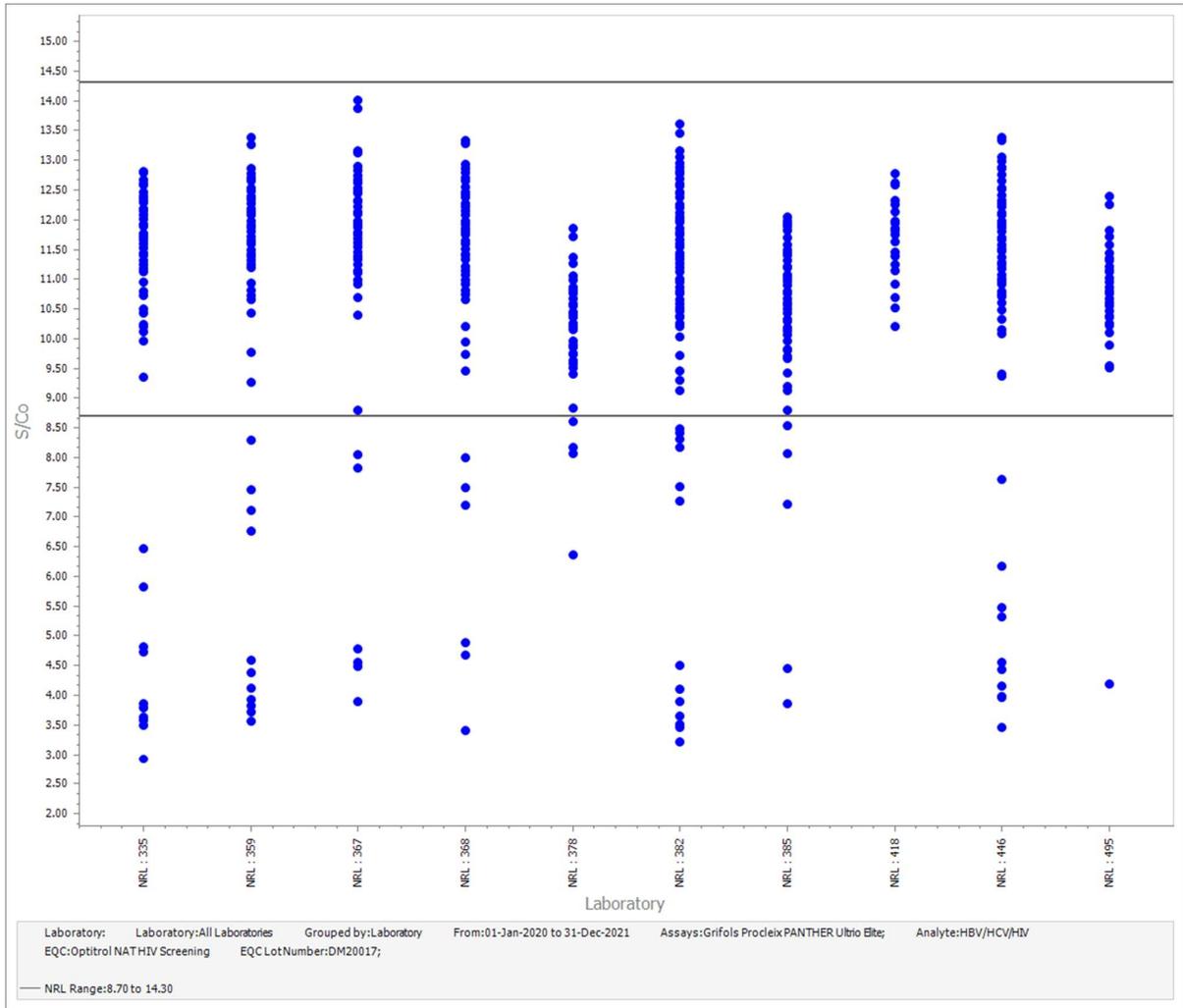


Figure 7 – Peer group data for Grifols Procleix PANTHER Ultrio Elite, Optitrol NAT HIV Screening DM20017 by participant.

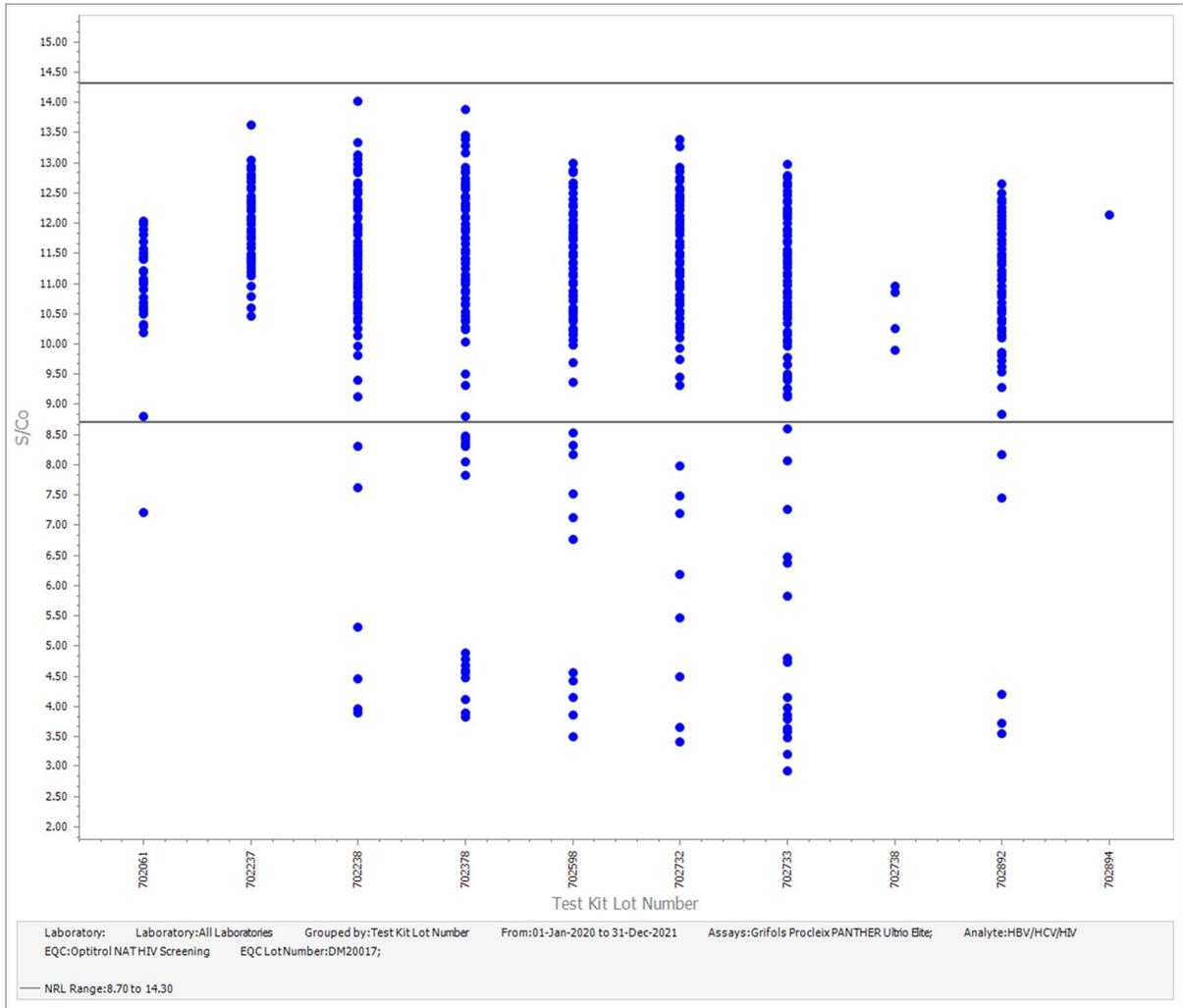


Figure 8 – Peer group data for Grifols Procleix PANTHER Ultrio Elite, Optitrol NAT HIV Screening DM20017 by reagent lot.

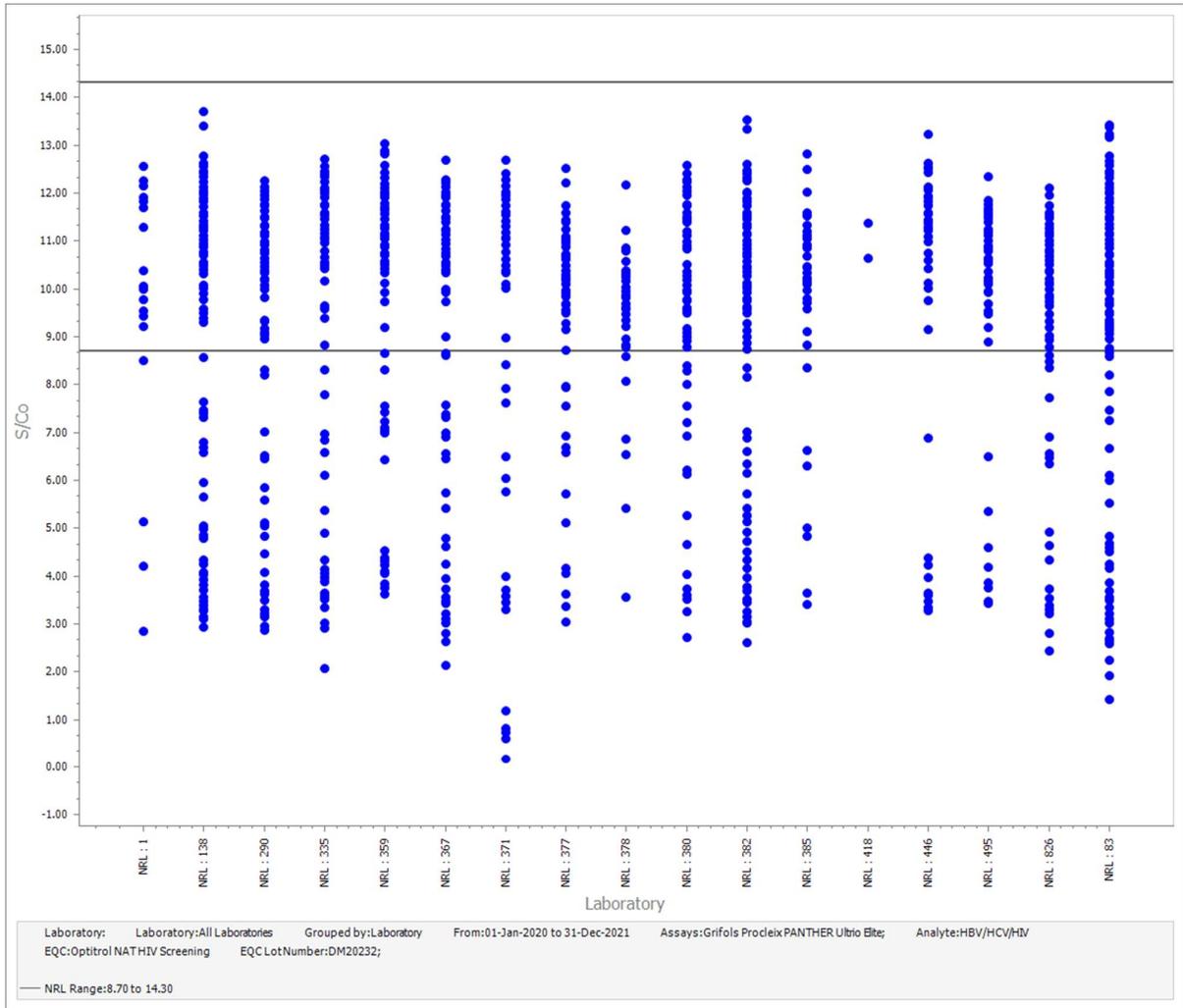


Figure 9 – Peer group data for Grifols Procleix PANTHER Ultrio Elite, Optitrol NAT HIV Screening DM20232 by participant.

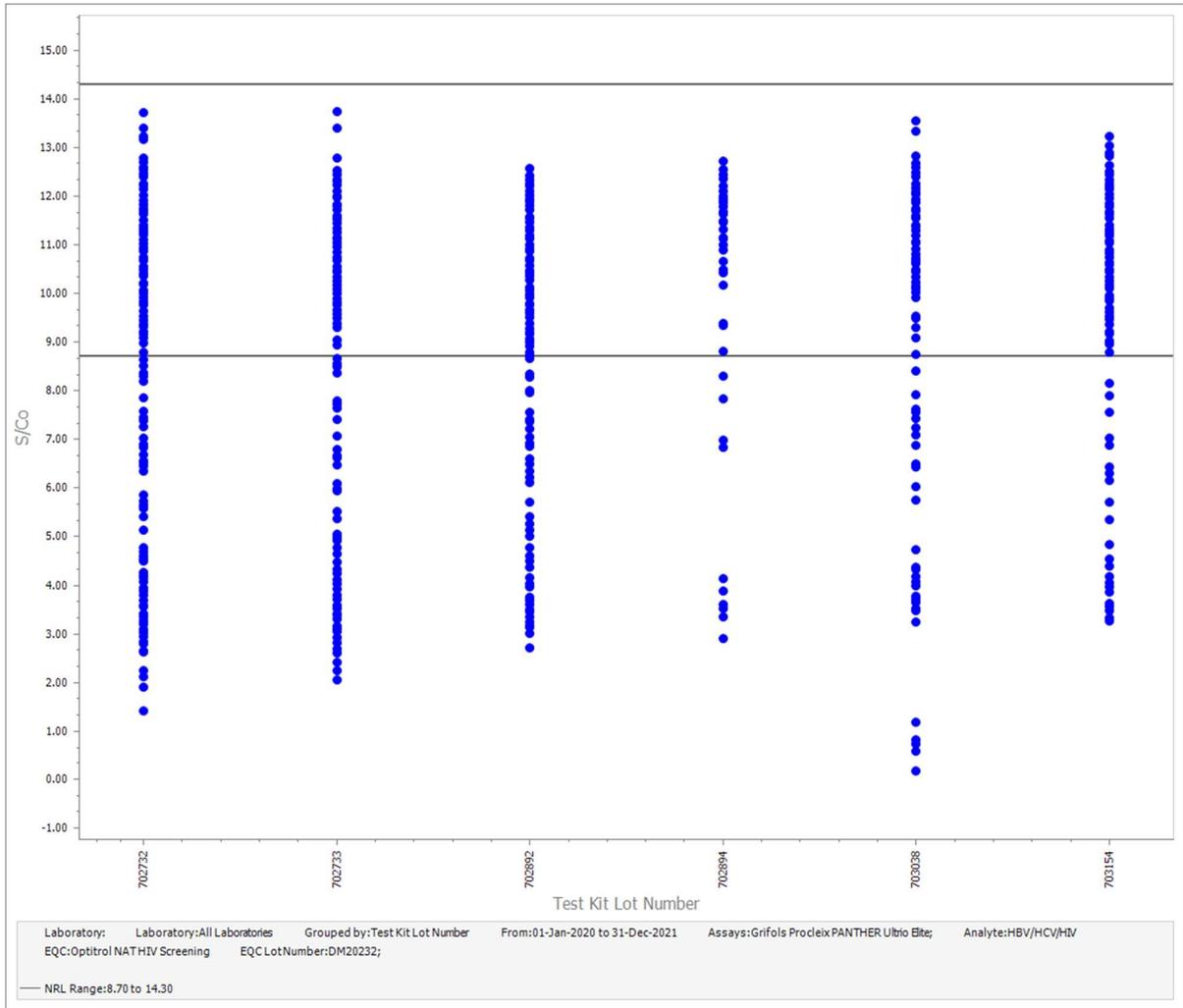


Figure 10 – Peer group data for Grifols Procleix PANTHER Ultrio Elite, Optitrol NAT HIV Screening DM20232 by reagent lot.

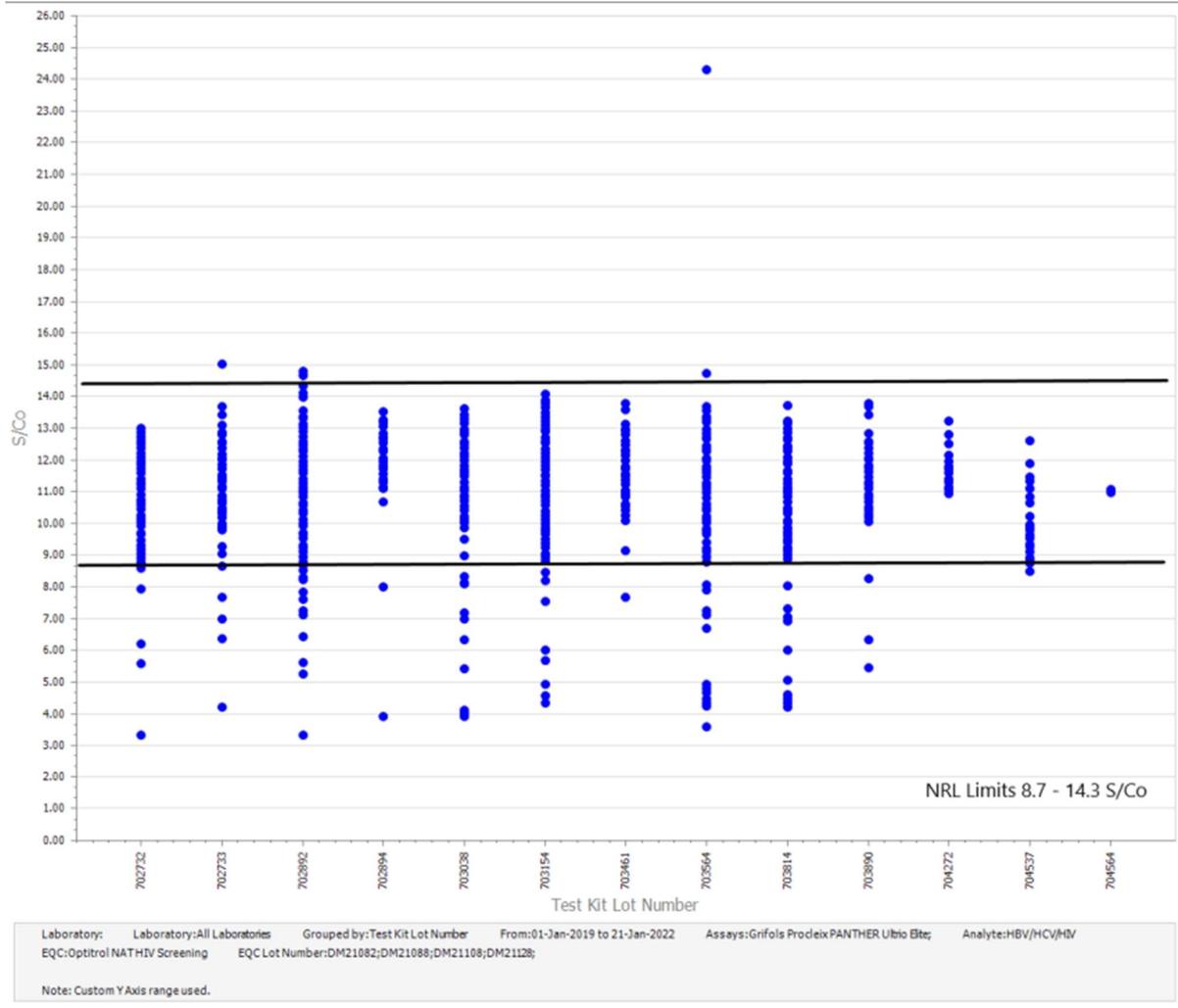


Figure 11 - Peer group data for Grifols Procleix PANTHER Ultra Elite, Optitrol NAT HIV Screening by reagent lot.

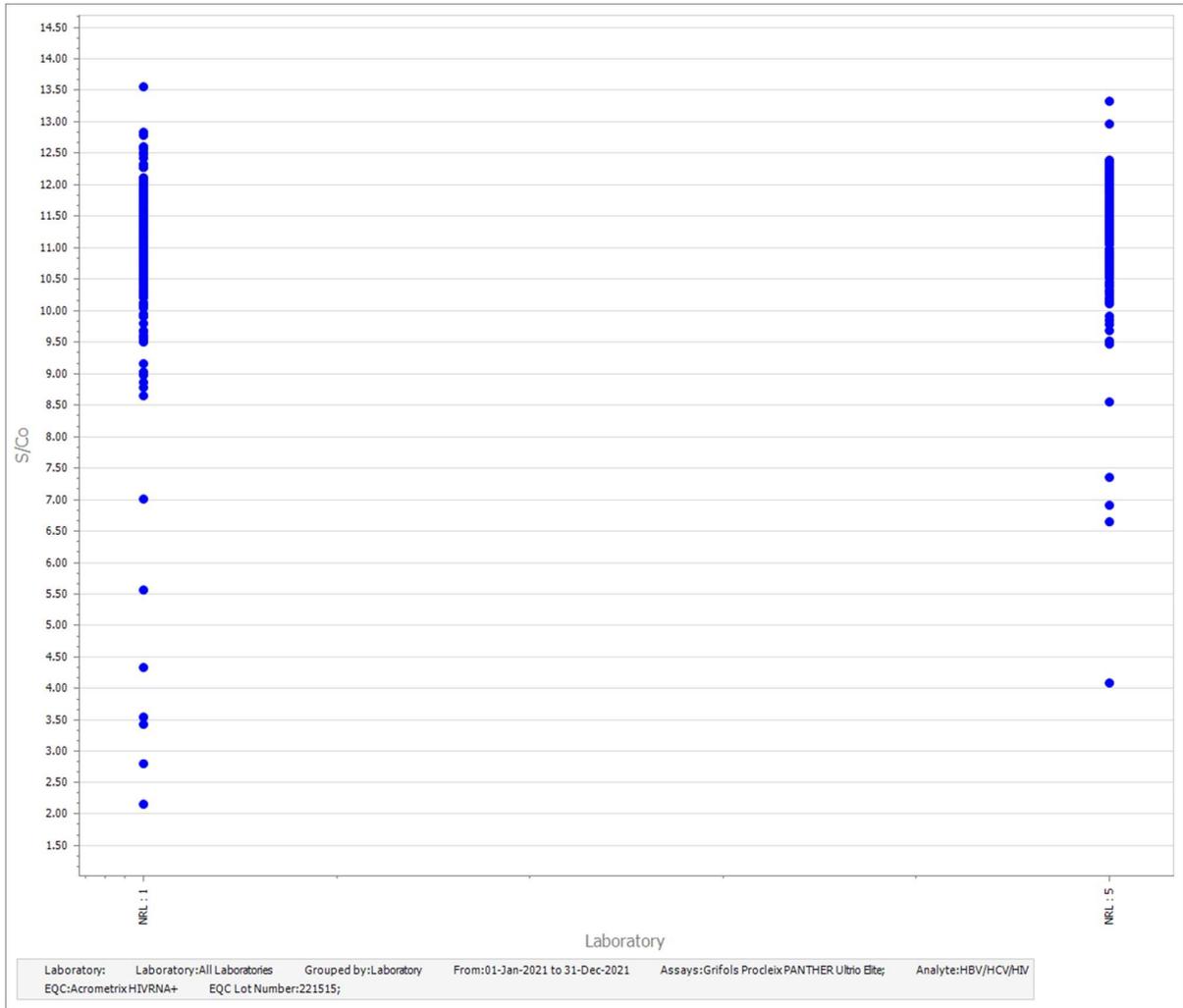


Figure 12 – Peer group data for Grifols Procleix PANTHER Ultrio Elite, AcroMetrix HIVRNA+ 221515 by participant.

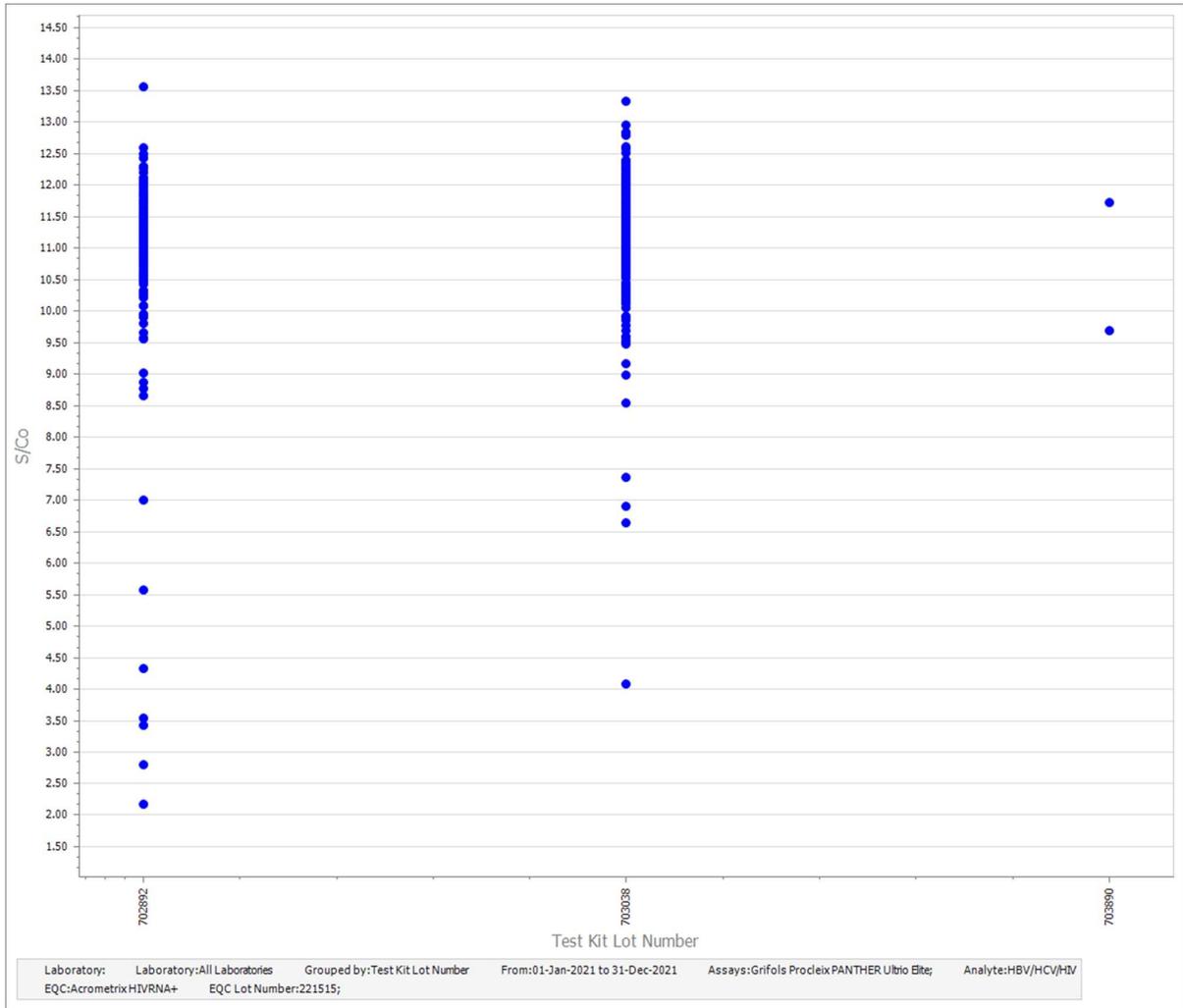


Figure 13 – Peer group data for Grifols Procleix PANTHER Ultrio Elite, AcroMetrix HIVRNA+ 221515 by reagent lot.

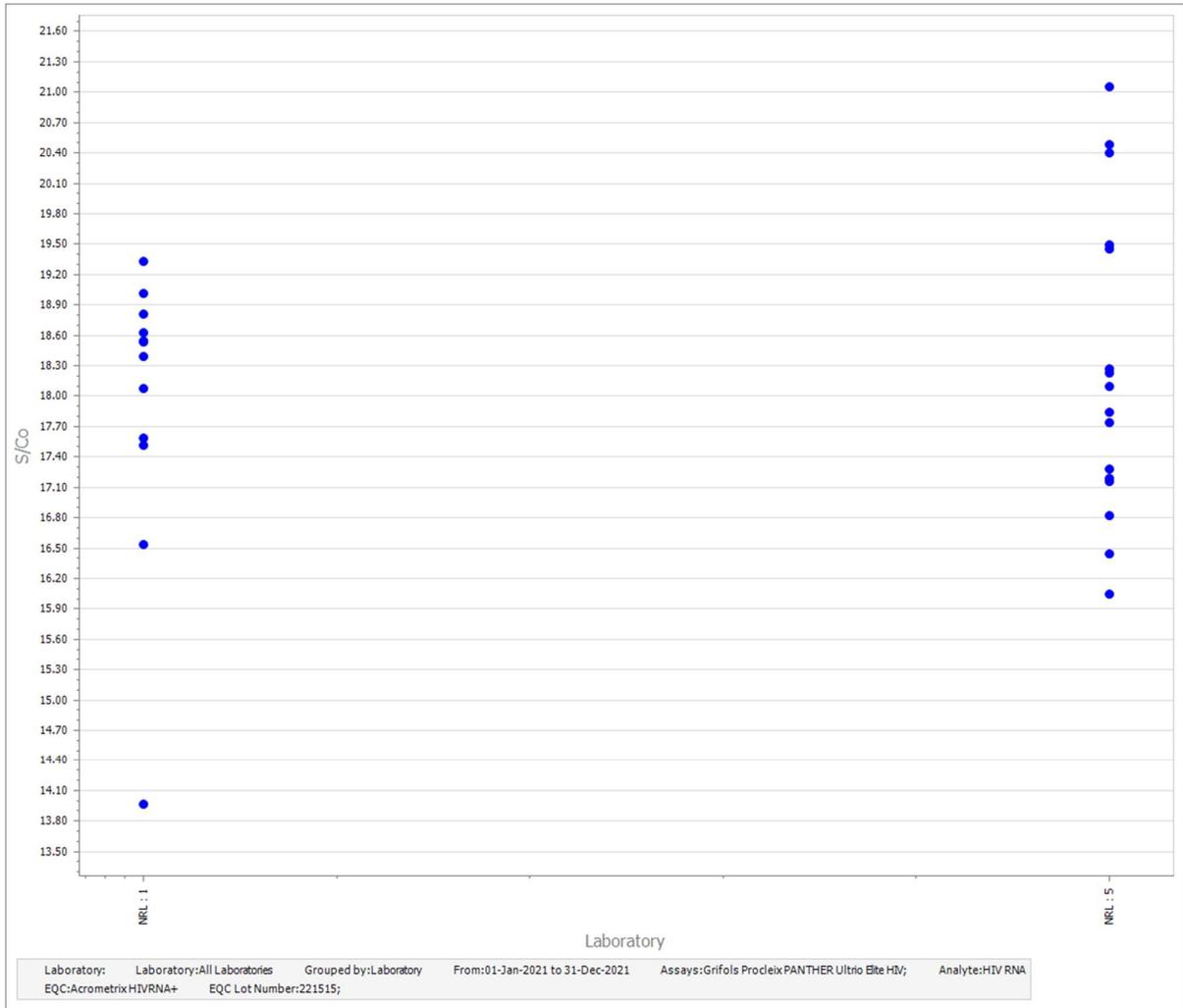


Figure 14 – Peer group data for Grifols Procleix PANTHER Ultrio Elite HIV, AcroMetrix HIVRNA+ 221515 by participant.

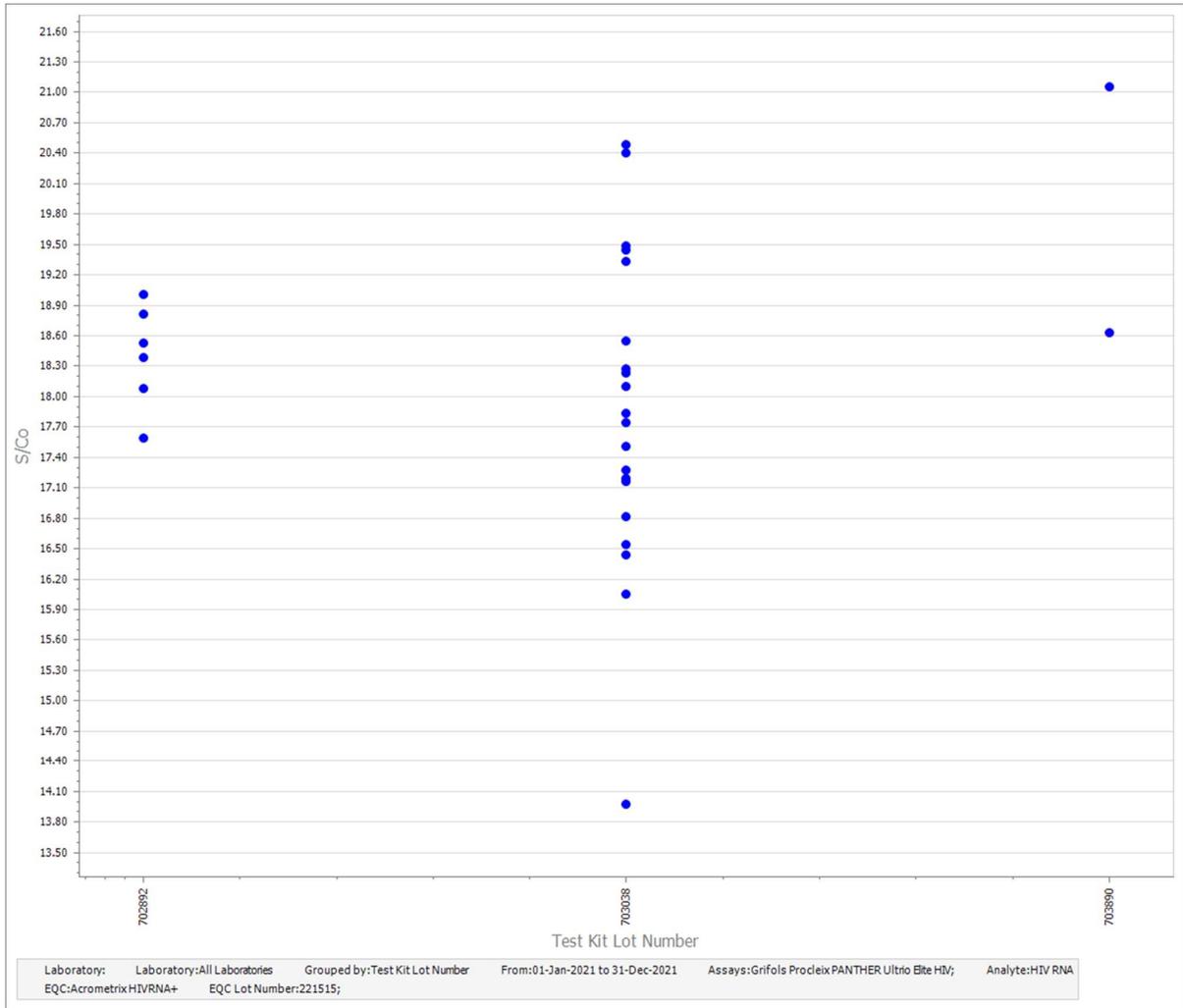


Figure 15 – Peer group data for Grifols Procleix PANTHER Ultrio Elite HIV, AcroMetrix HIVRNA+ 221515 by reagent lot.

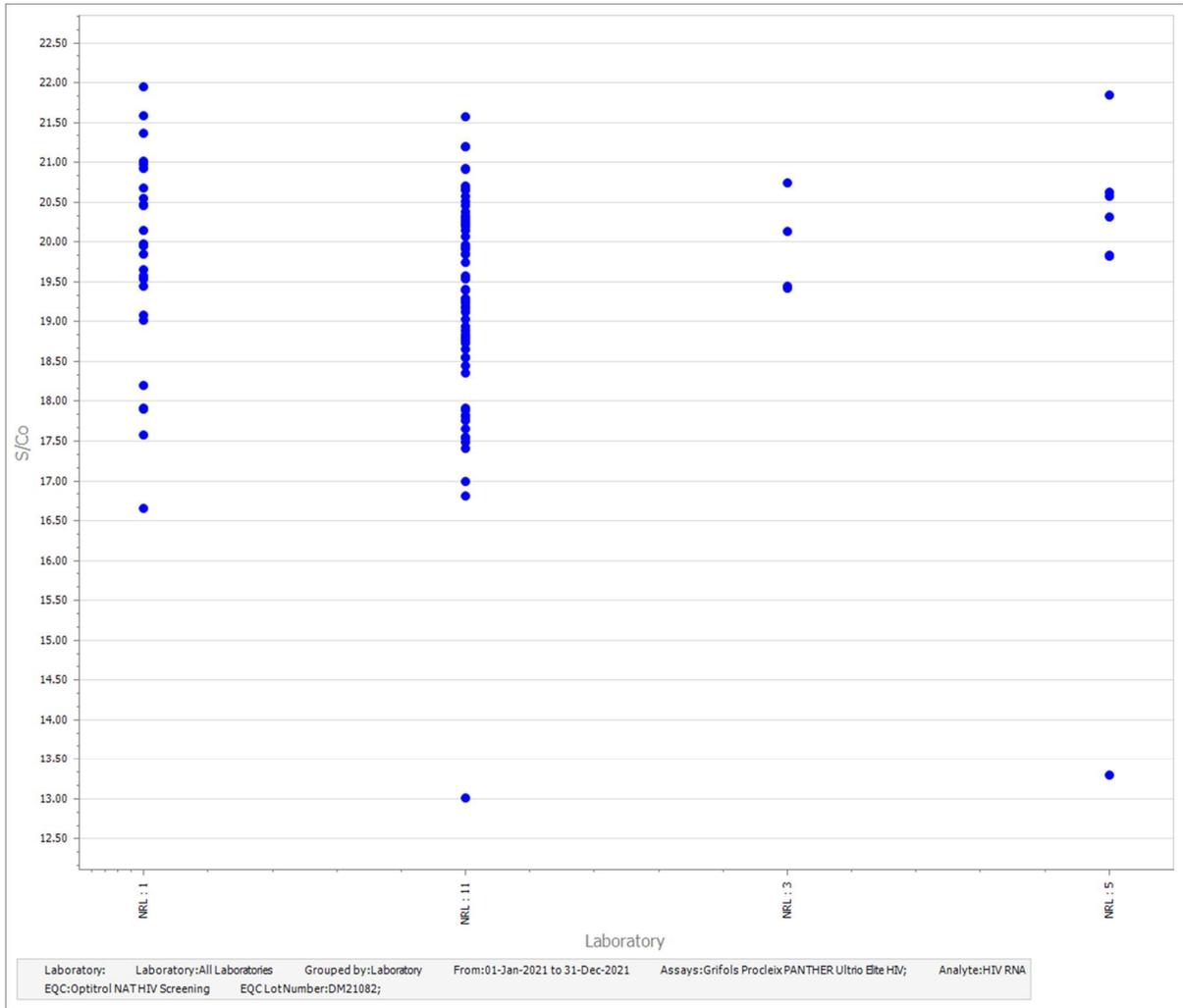


Figure 16 – Peer group data for Grifols Procleix PANTHER Ultrio Elite HIV, Optitrol NAT HIV Screening DM21082 by participant.

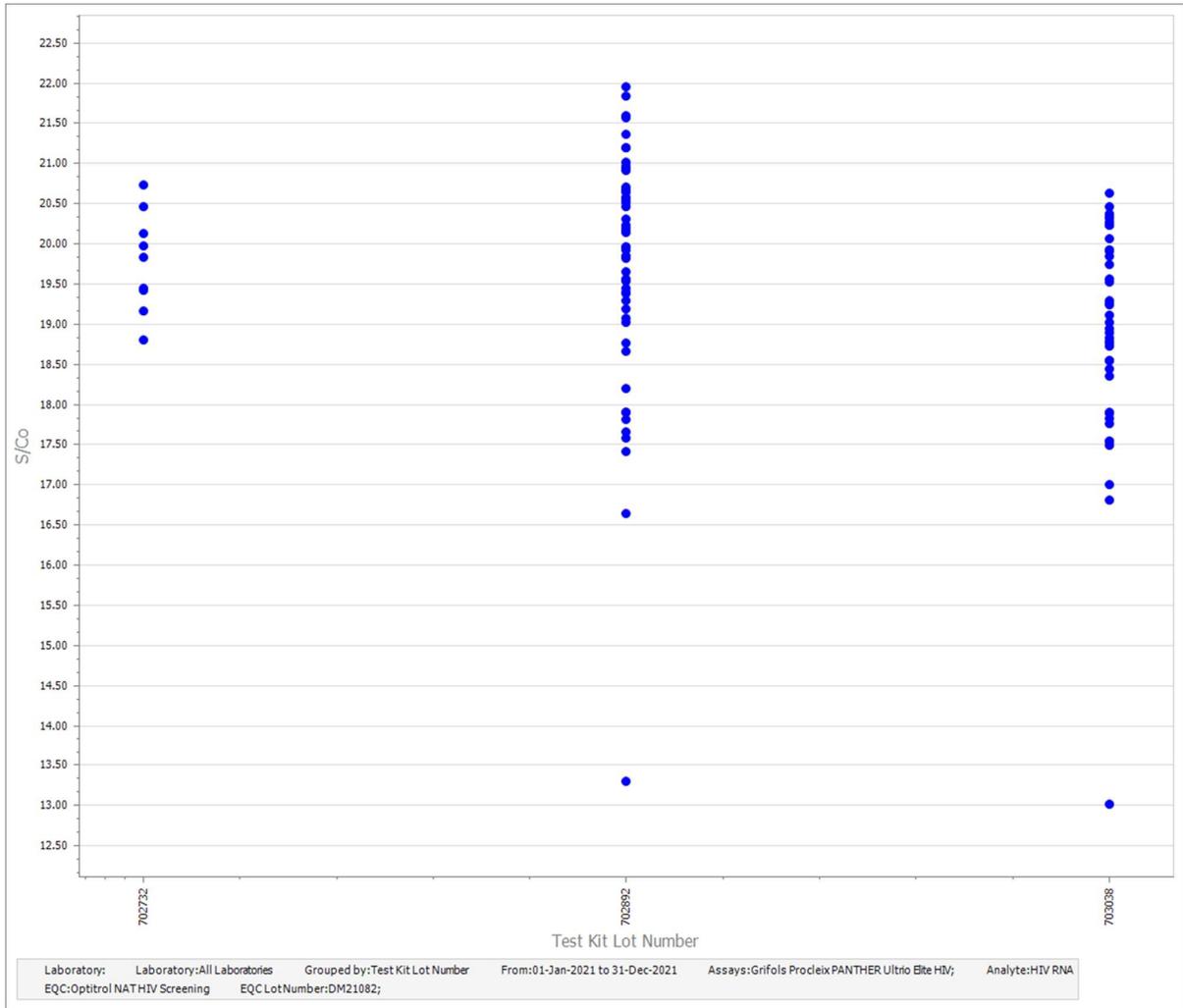
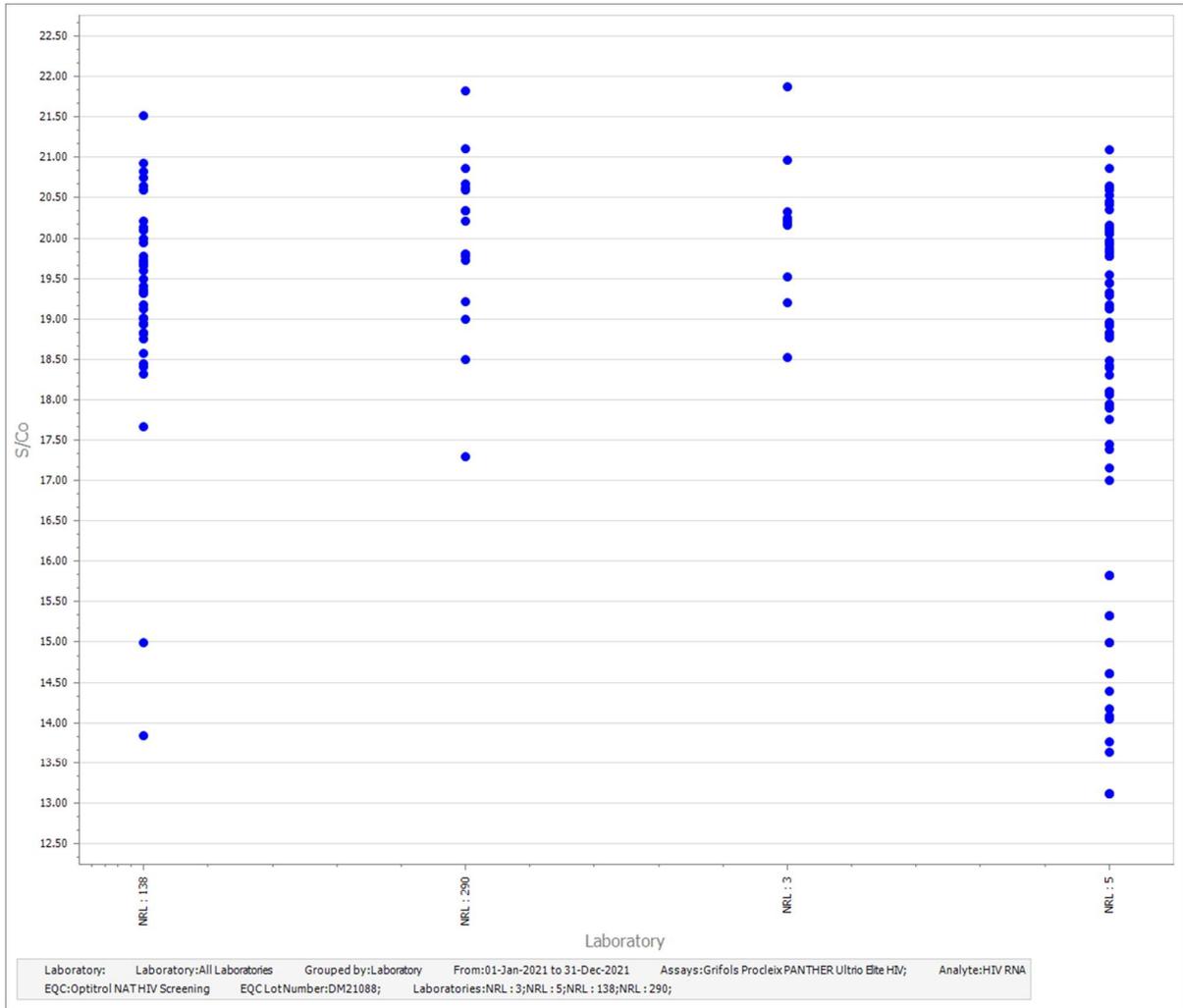


Figure 17 – Peer group data for Grifols Procleix PANTHER Ultrio Elite HIV, Optitrol NAT HIV Screening DM21082 by reagent lot.



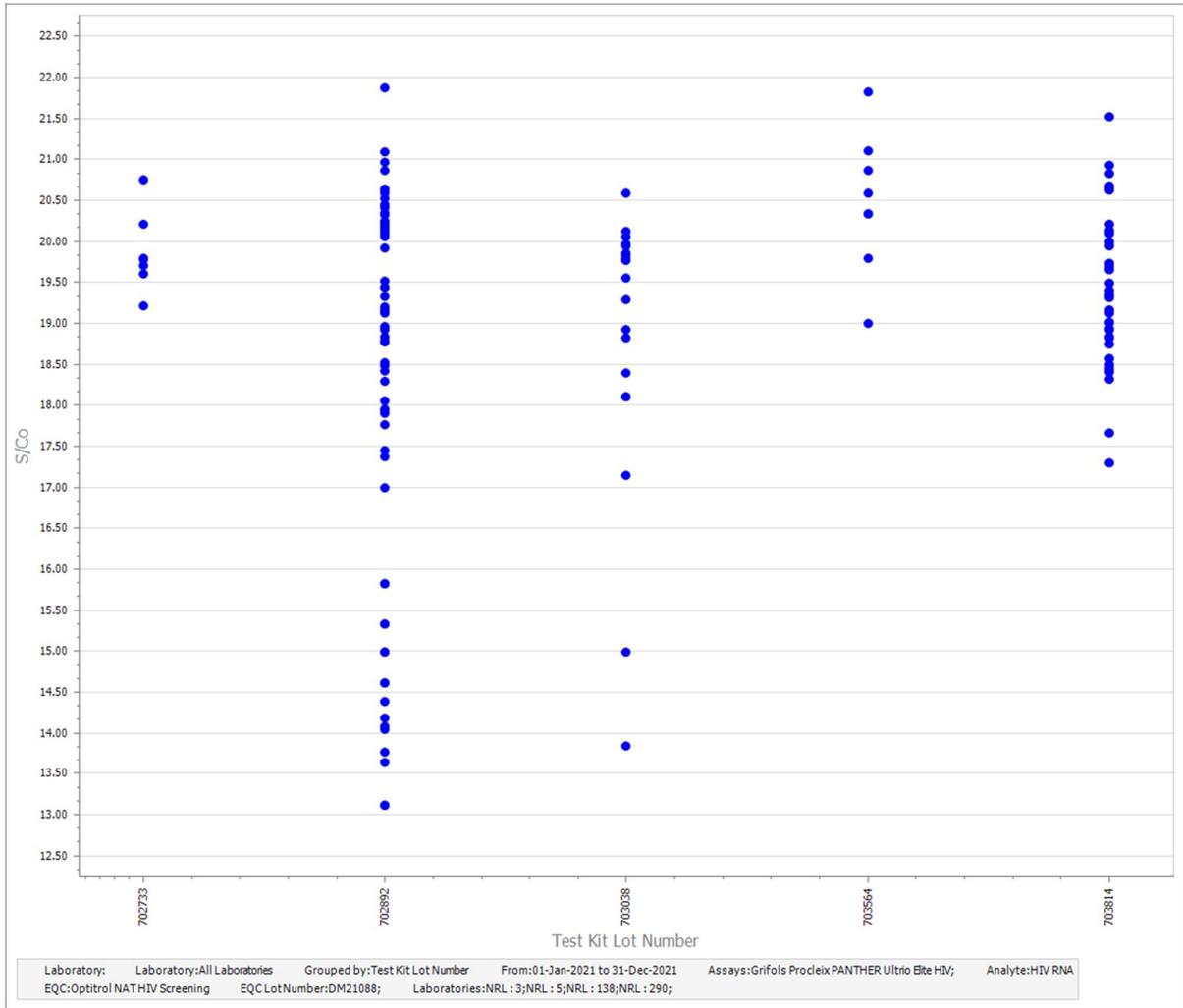


Figure 19 – Peer group data for Grifols Procleix PANTHER Ultrio Elite HIV, Optitrol NAT HIV Screening DM21088 by reagent lot.

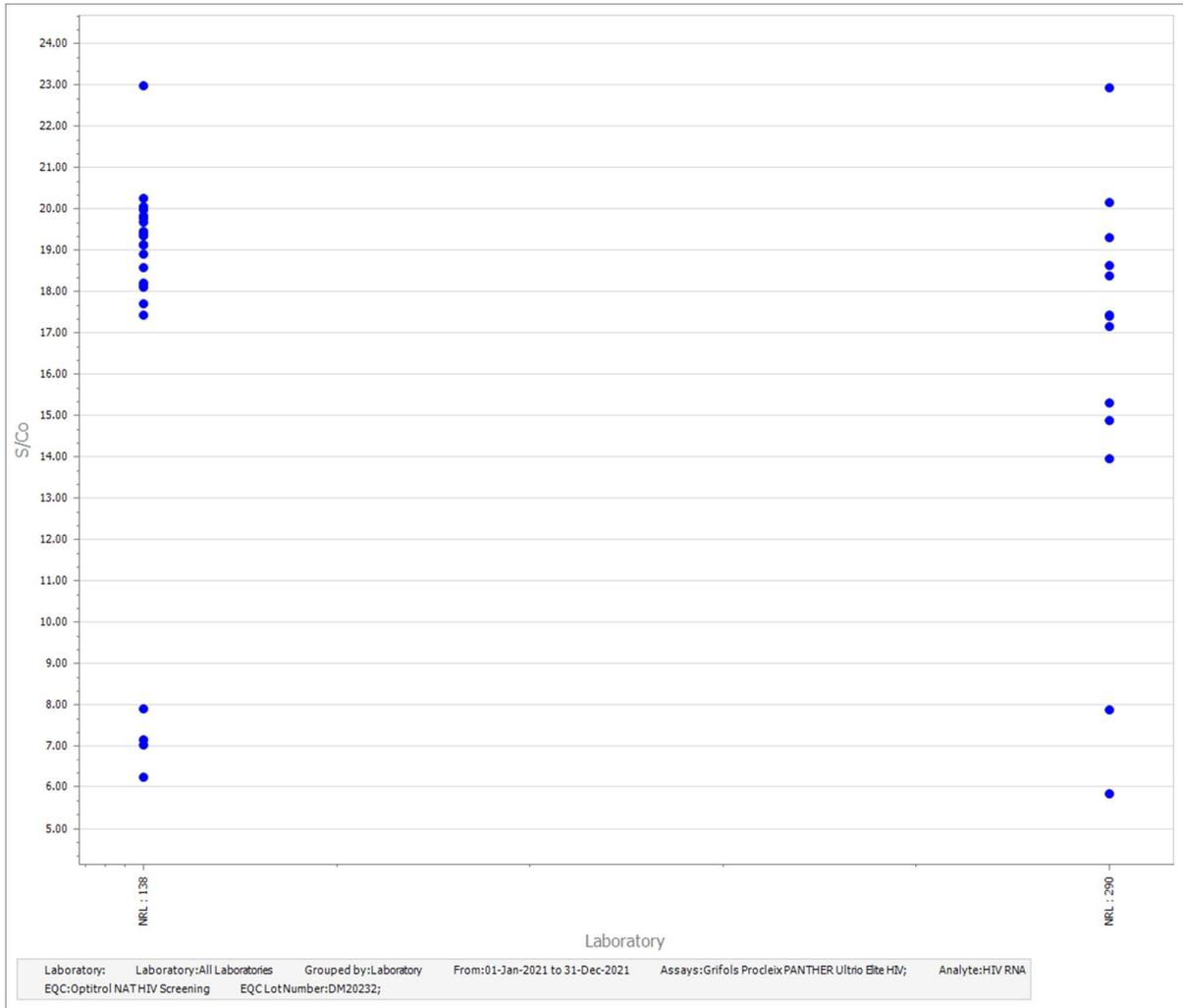


Figure 20 – Peer group data for Grifols Procleix PANTHER Ultrio Elite HIV, Optitrol NAT HIV Screening DM20232 by participant.

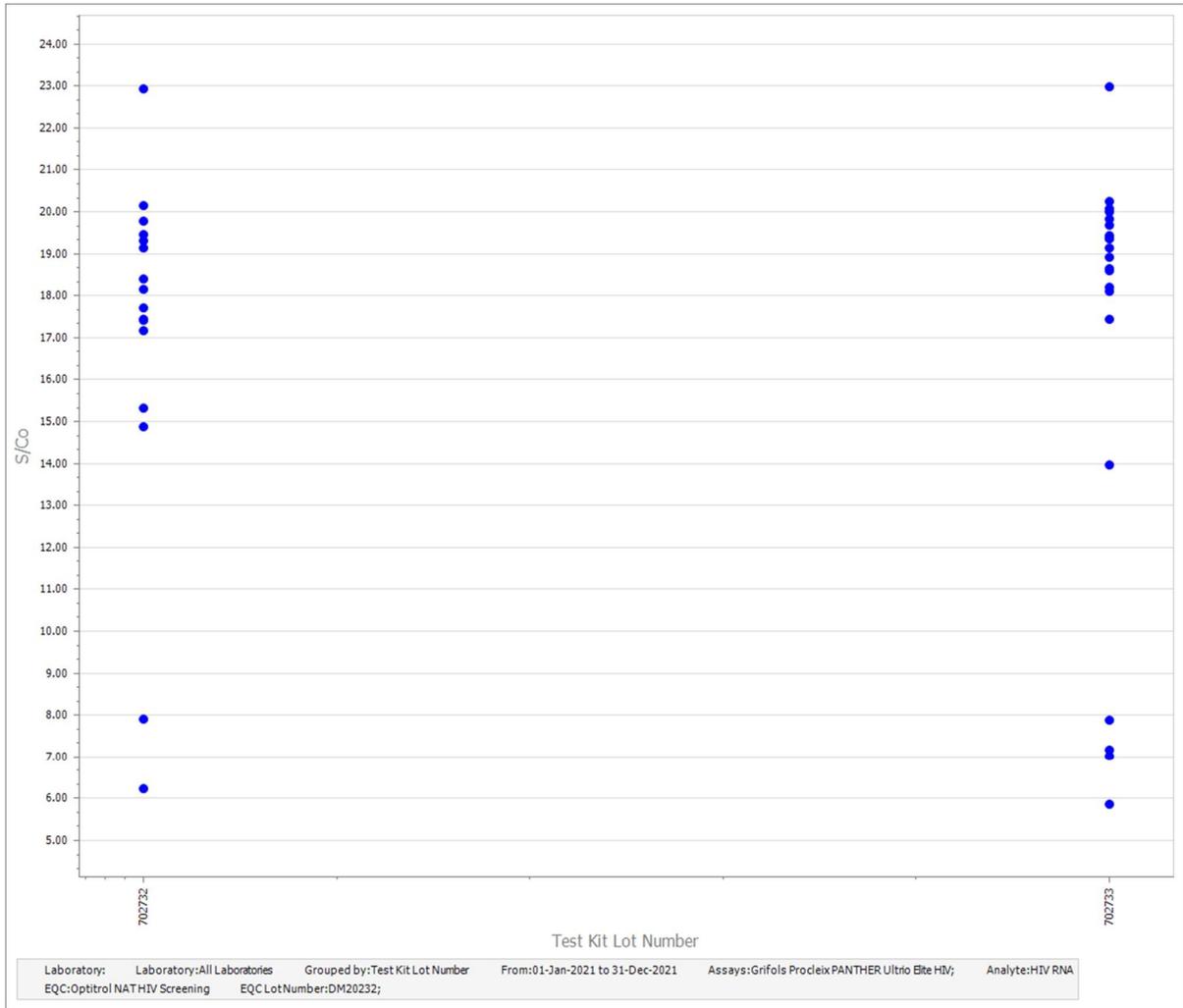


Figure 21 – Peer group data for Grifols Procleix PANTHER Ultrio Elite HIV, Optitrol NAT HIV Screening DM20232 by reagent lot.

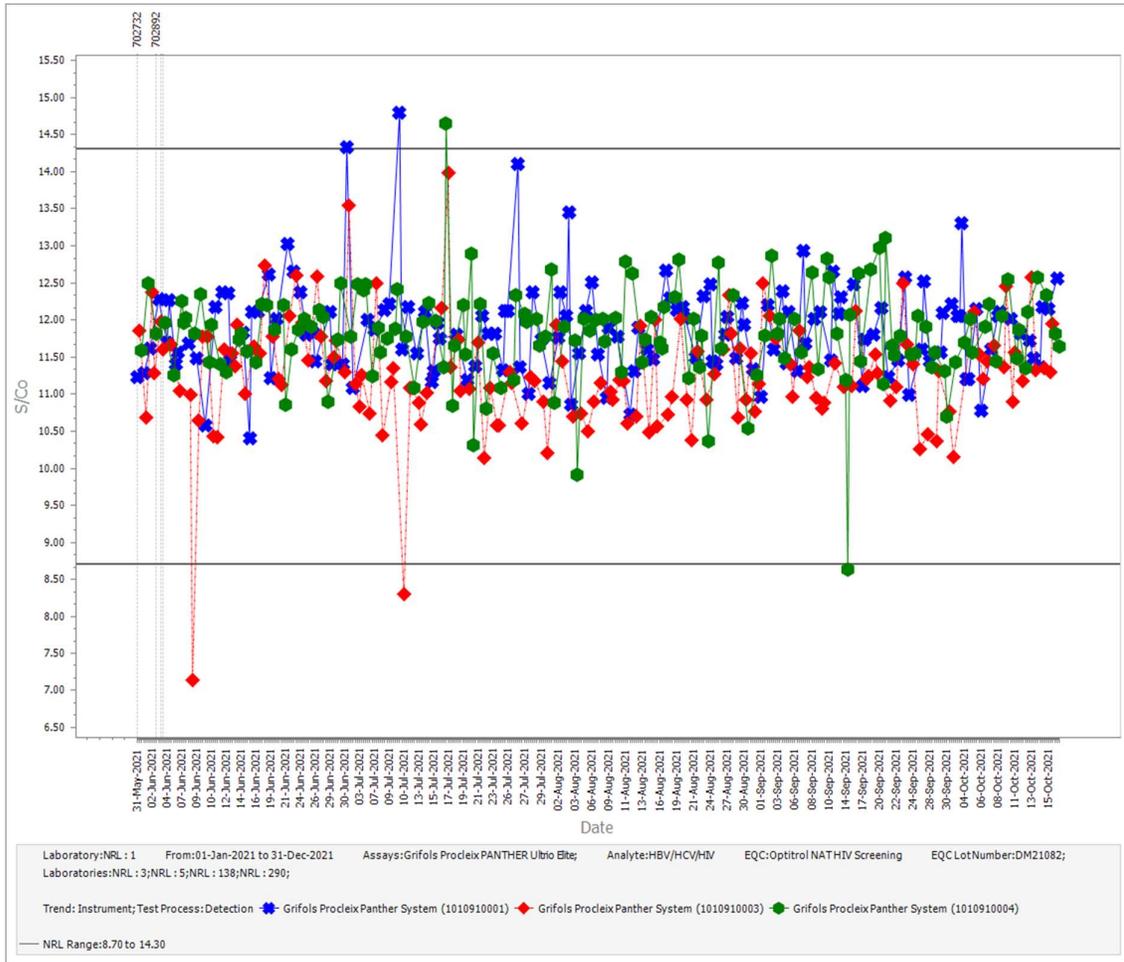


Figure 22 - Participant 1 data for Grifols Procleix PANTHER Ultrio Elite, Optitrol NAT HIV Screening DM21082 by instrument.

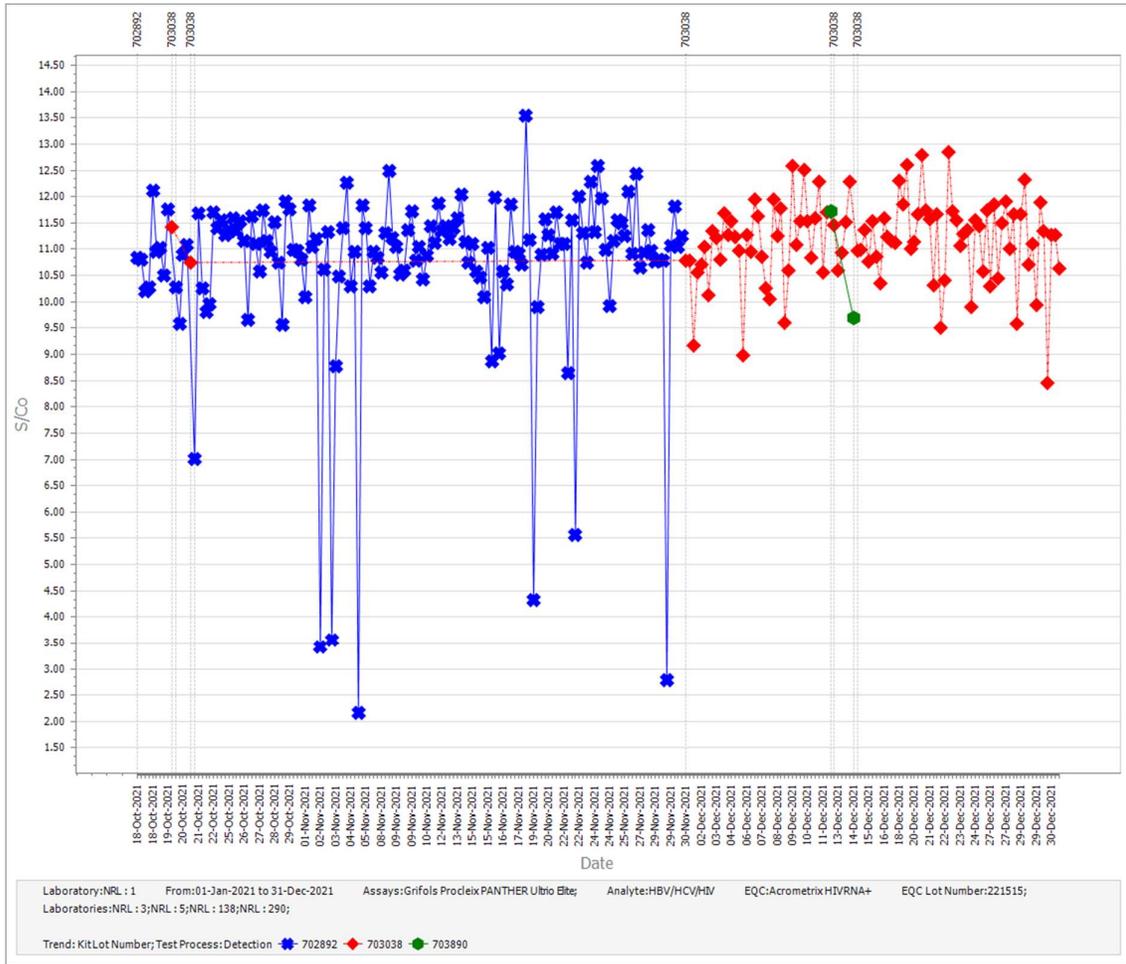


Figure 23 - Participant 1 data for Grifols Procleix PANTHER Ultrio Elite, AcroMetrix HIVRNA+ 221515 by reagent lot.

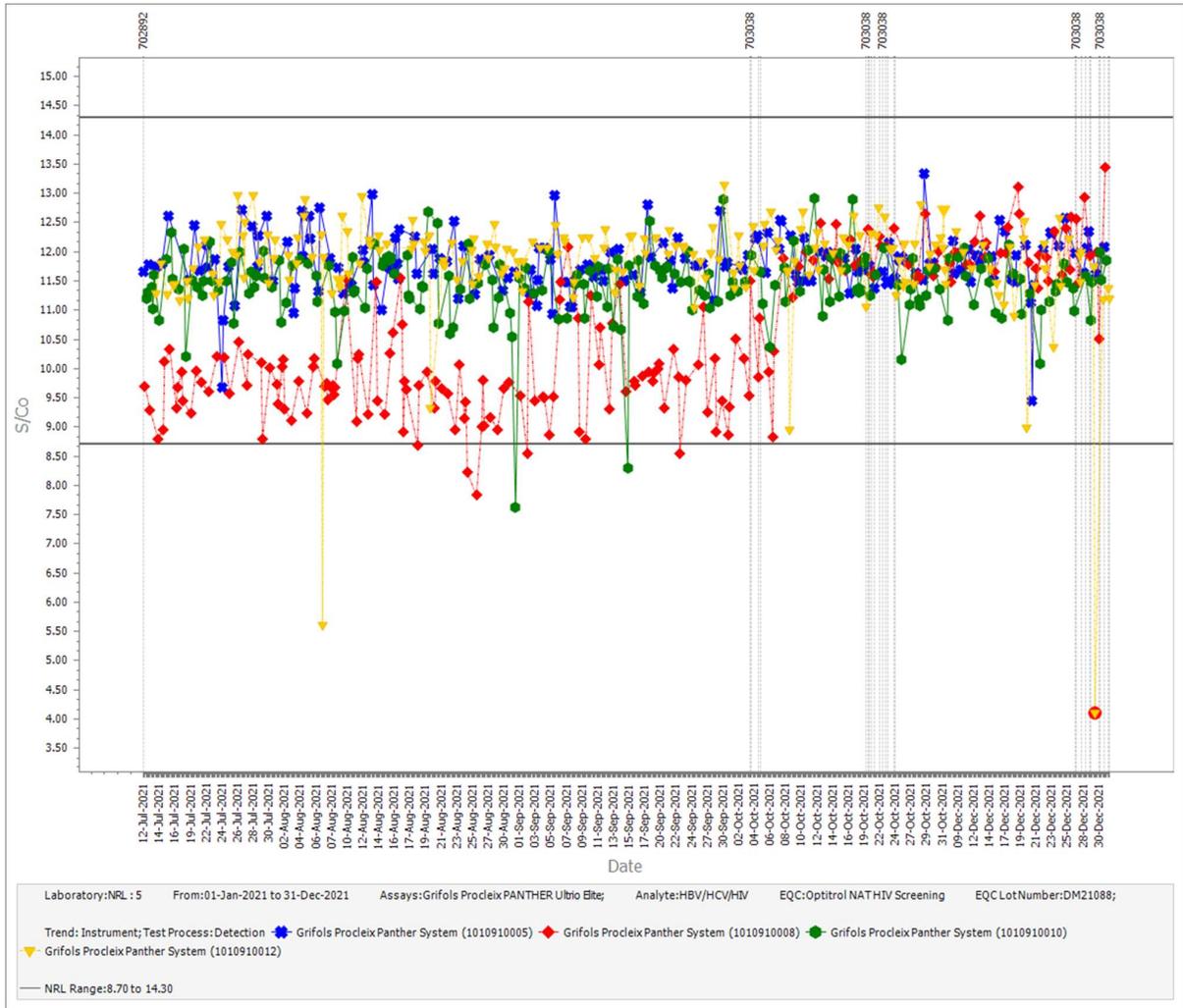


Figure 24 - Participant 5 data for Grifols Procleix PANTHER Ultrio Elite, Optitrol NAT HIV Screening DM21088 by instrument.

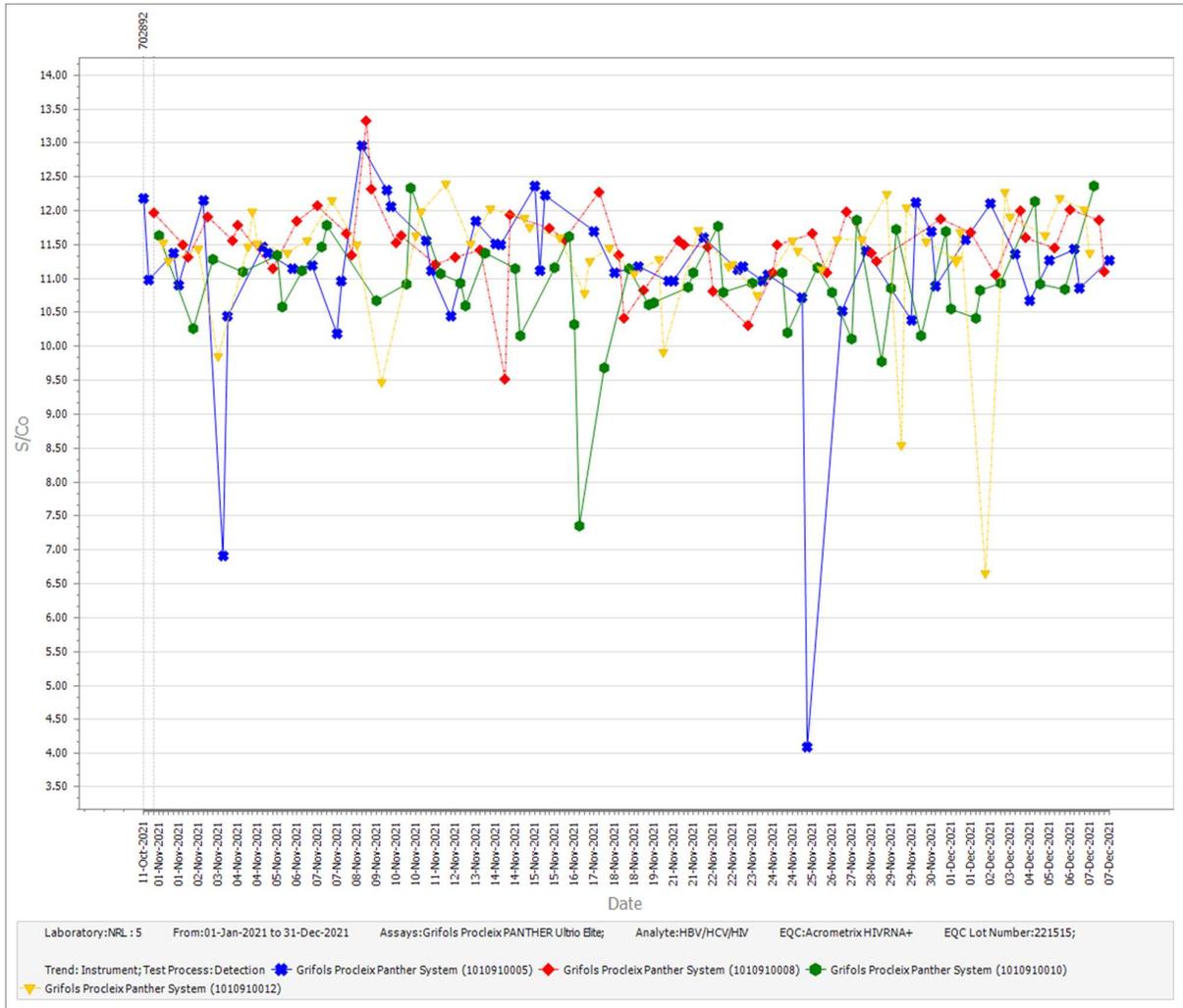


Figure 25 - Participant 5 data for Grifols Procleix PANTHER Ultrio Elite, AcroMetrix HIVRNA+ 221515 by instrument.

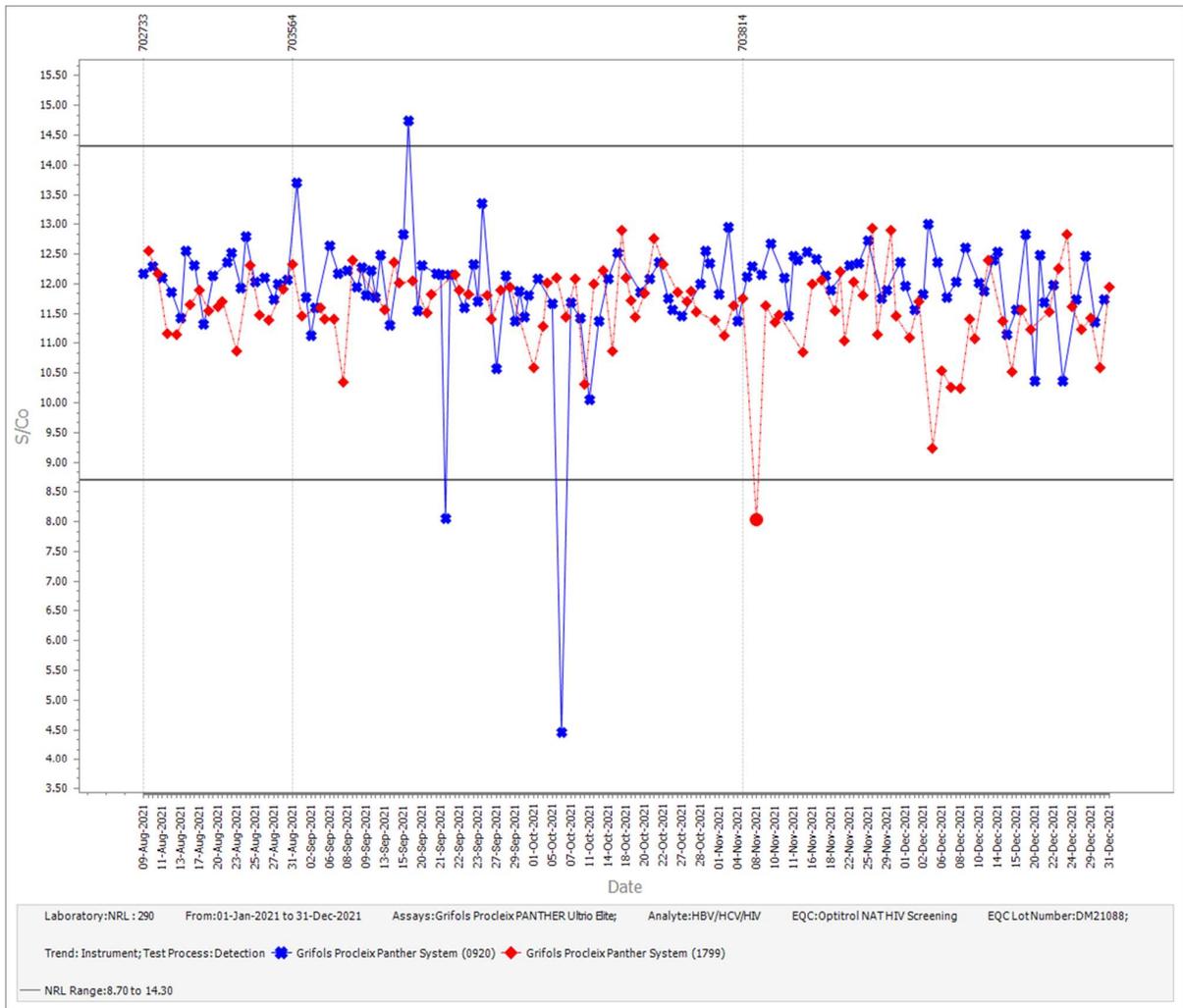


Figure 26 – Participant 290 data for Grifols Procleix PANTHER Ultrio Elite, Optitrol NAT HIV Screening DM21088 by instrument.

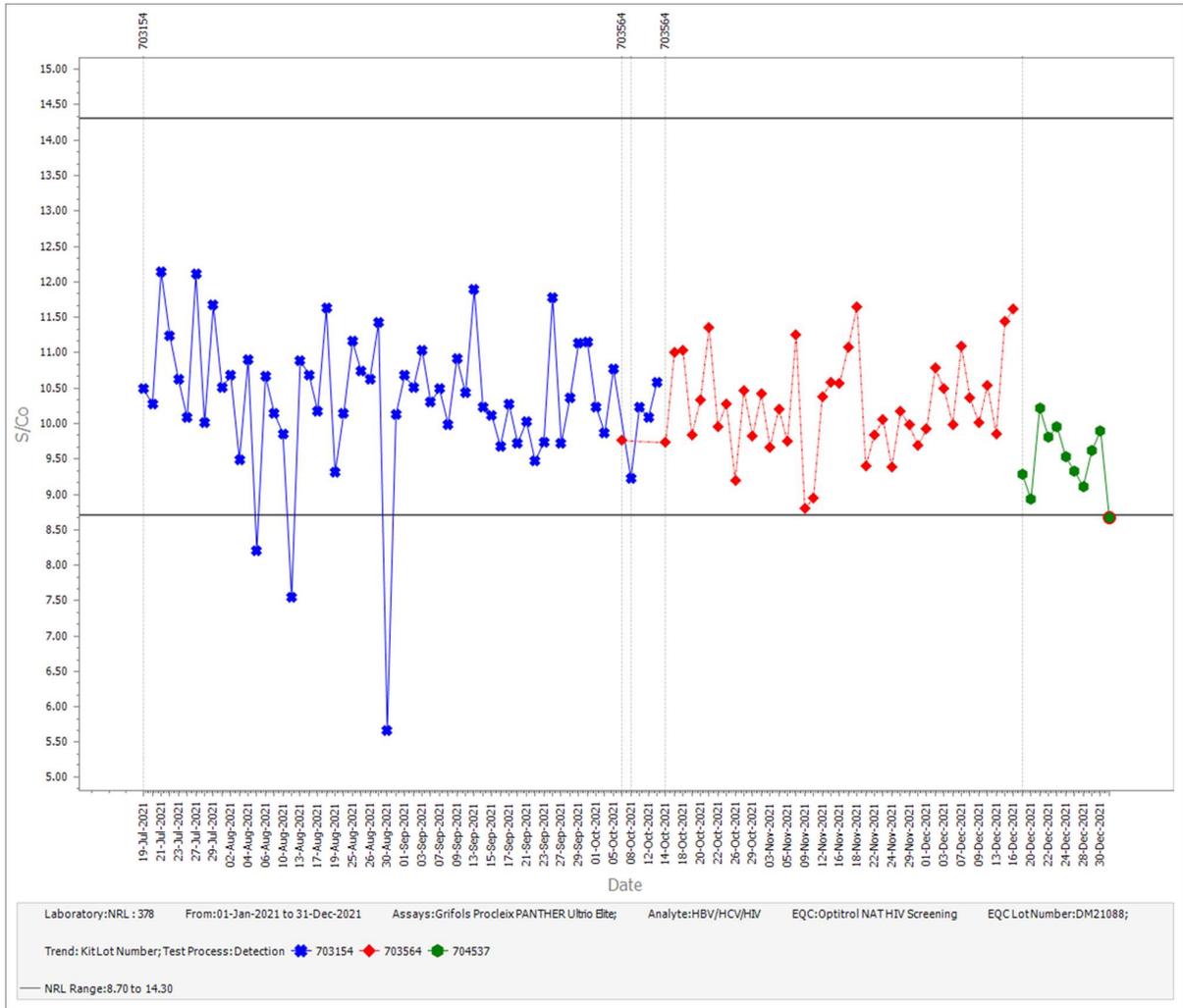


Figure 27 - Participant 378 data for Grifols Procleix PANTHER Ultrio Elite, Optitrol NAT HIV Screening DM21088 by reagent lot.

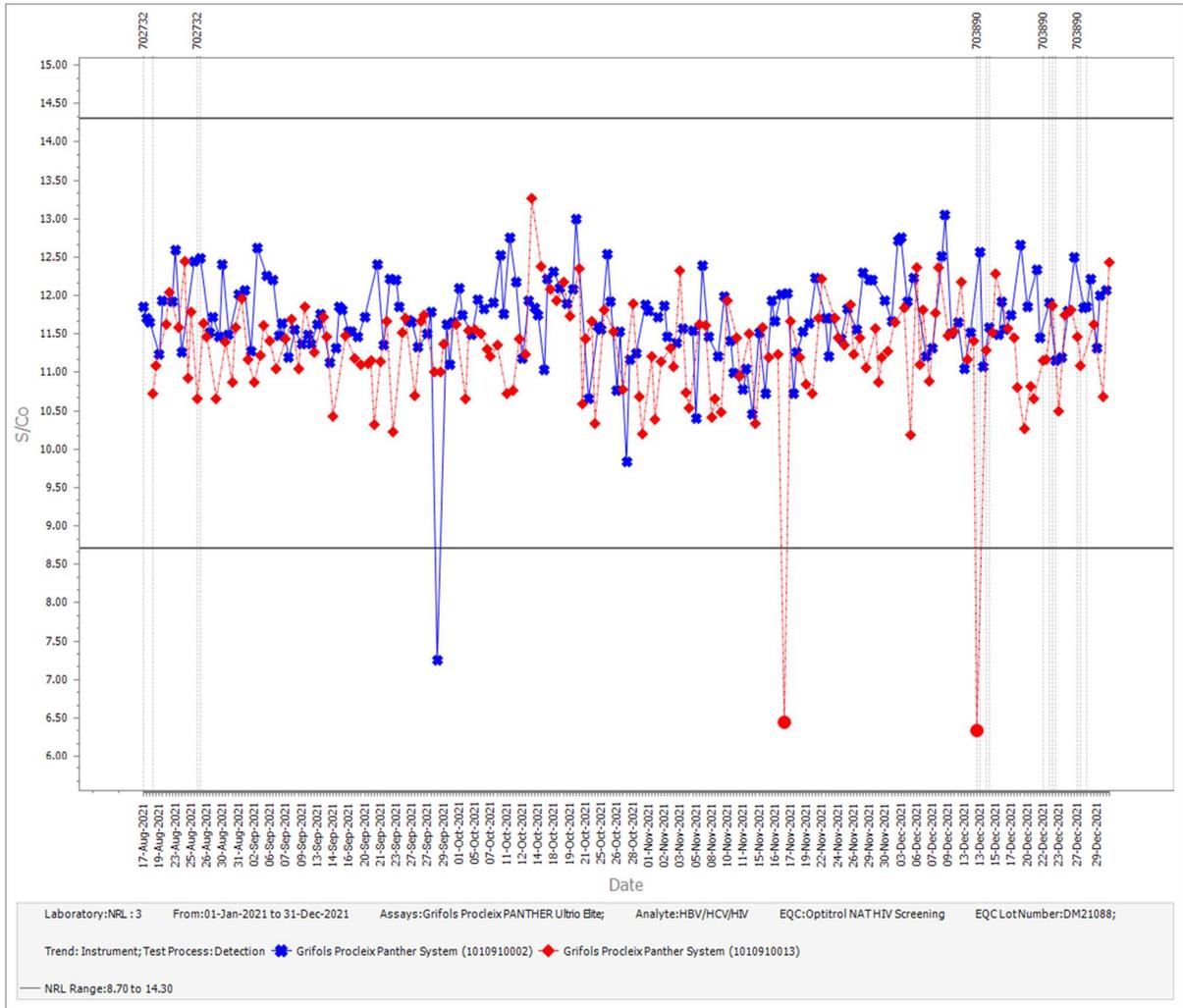


Figure 28 - Participant 3 data for Grifols Procleix PANTHER Ultrio Elite, Optitrol NAT HIV Screening DM21088 by instrument.

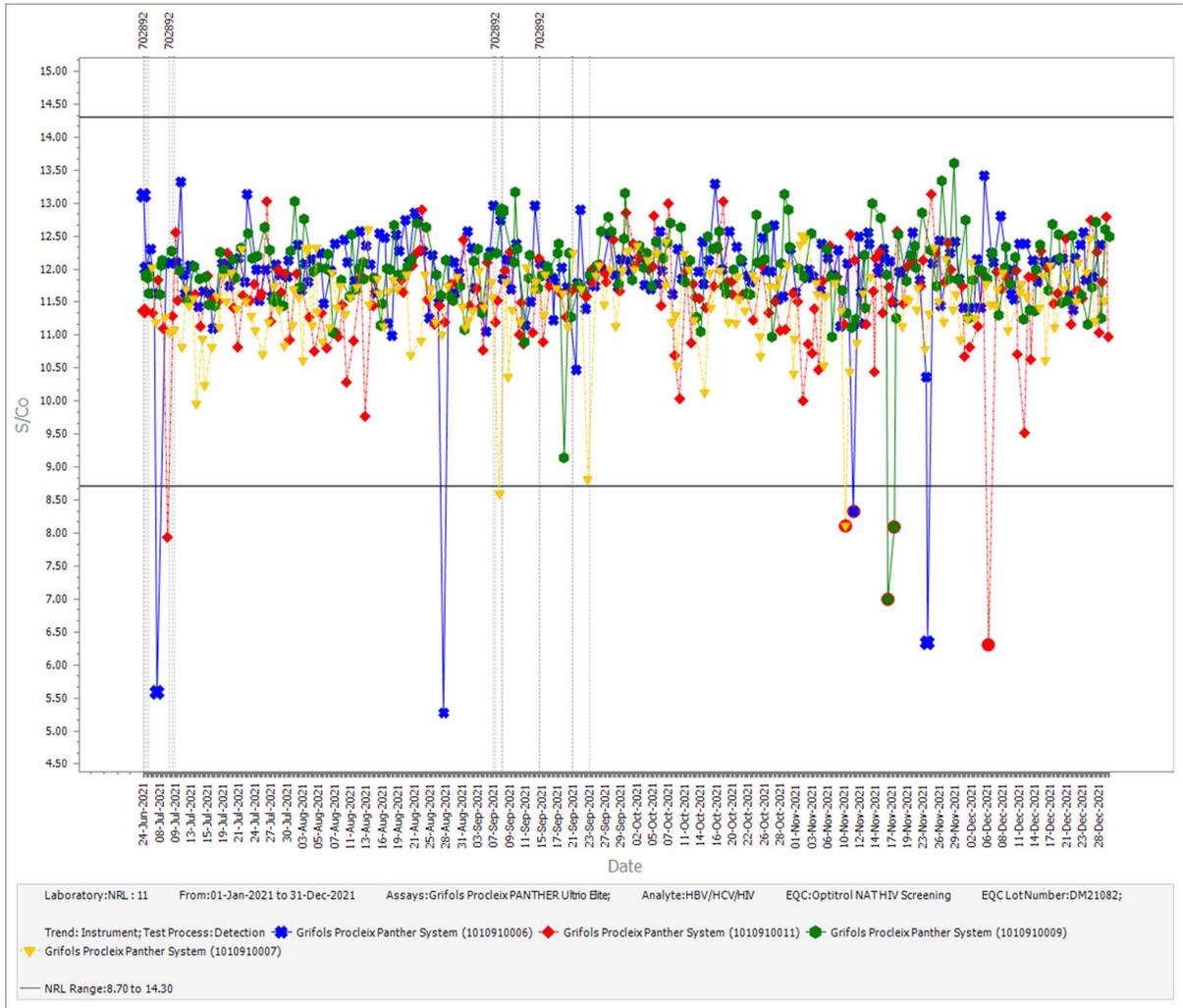


Figure 29 - Participant 11 data for Grifols Procleix PANTHER Ultrio Elite, Optitrol NAT HIV Screening DM21082 by instrument.

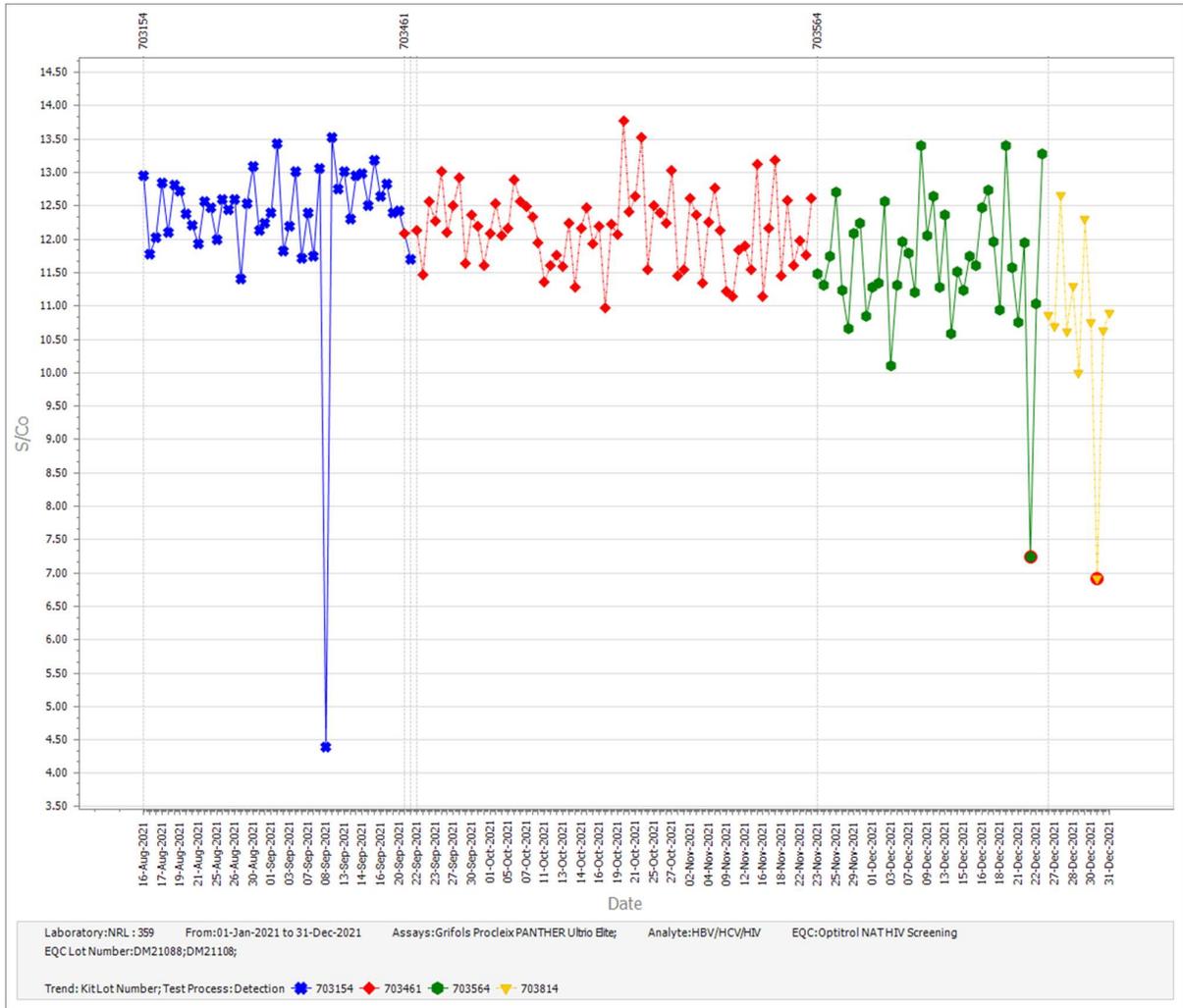


Figure 30 - Participant 359 data for Grifols Procleix PANTHER Ultrio Elite, Optitrol NAT HIV Screening DM21088 and DM21108 by reagent lot.

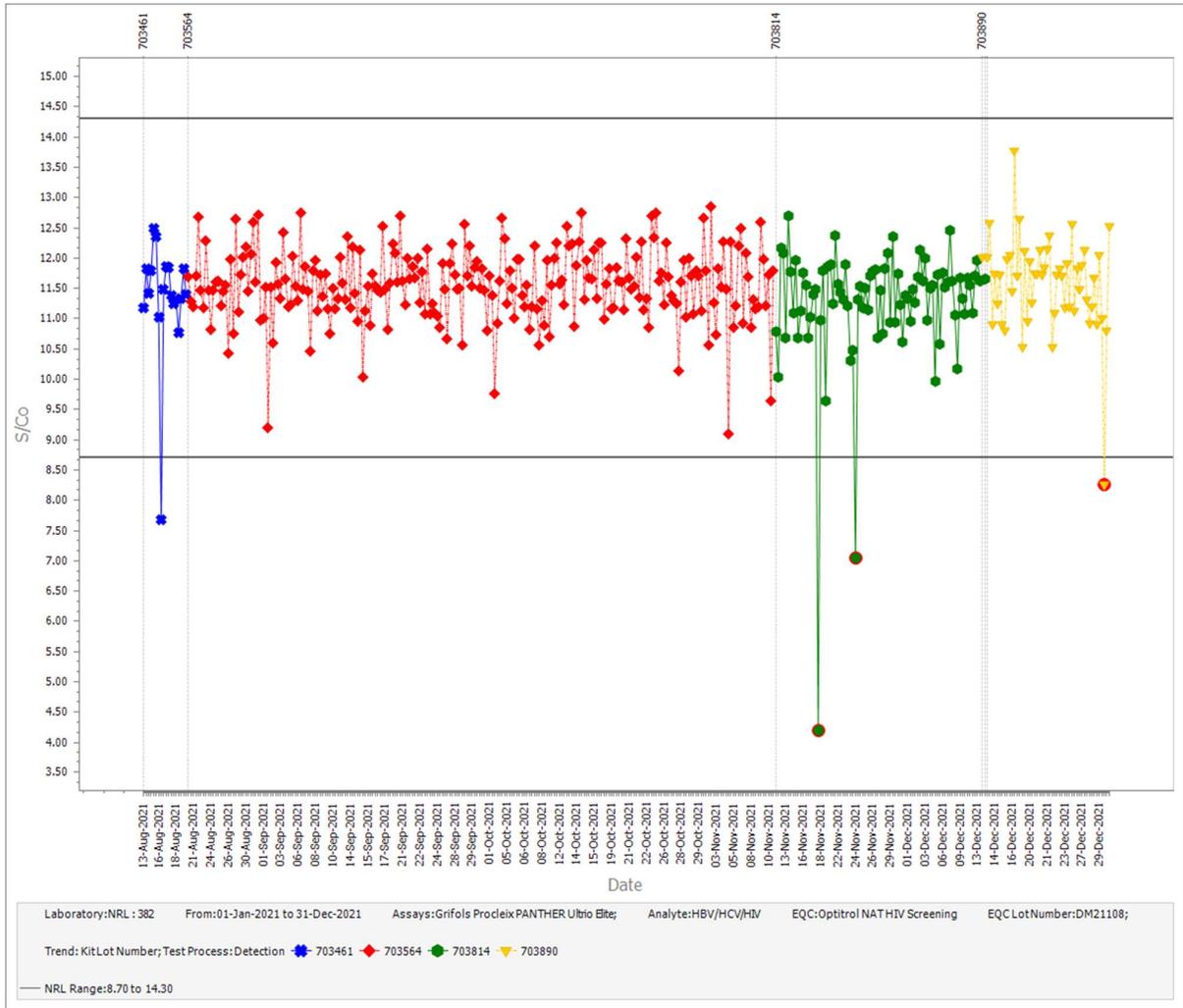


Figure 31 - Participant 382 data for Grifols Procleix PANTHER Ultrio Elite, Optitrol NAT HIV Screening DM21108 by reagent lot.

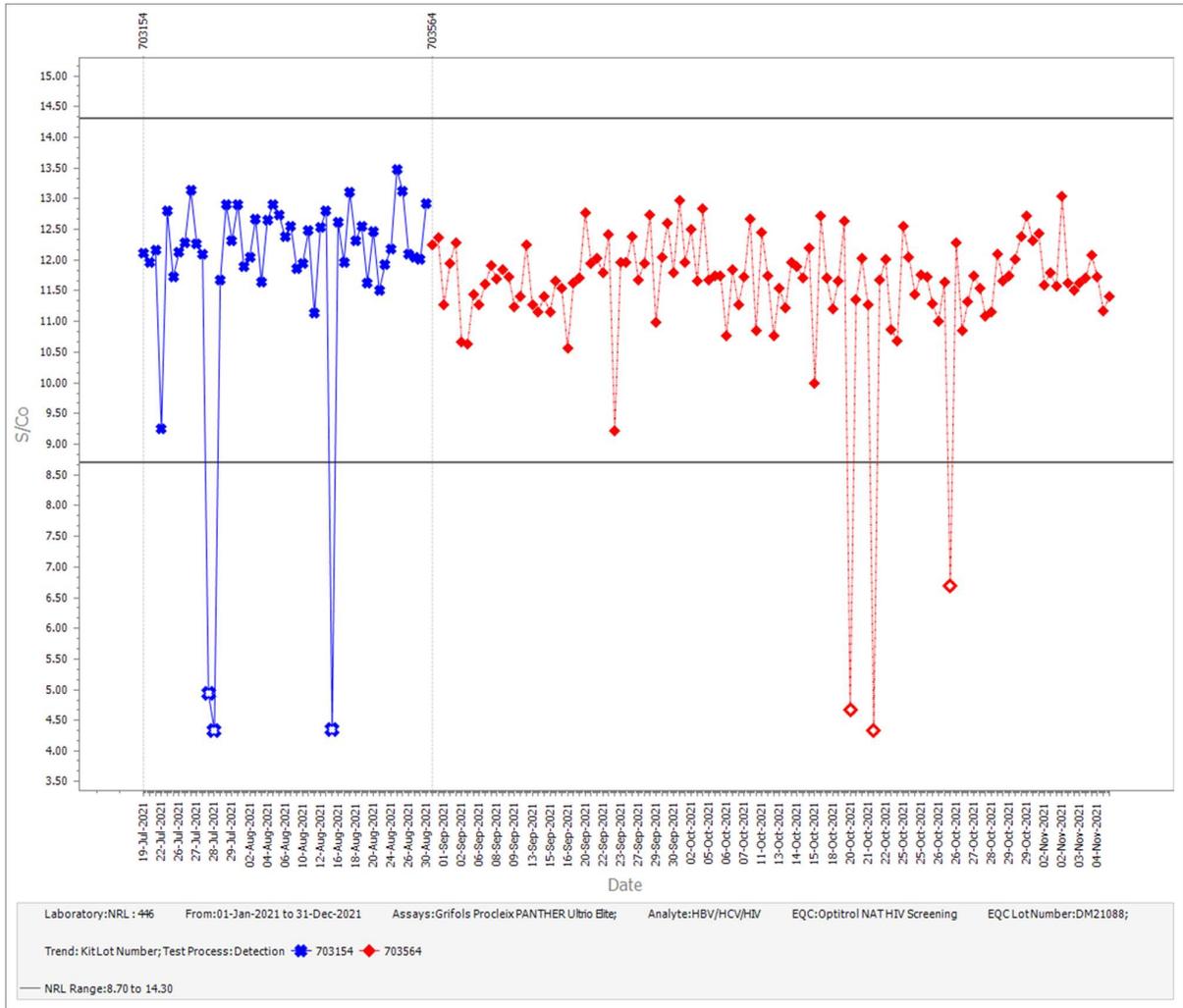


Figure 32 - Participant 446 data for Grifols Procleix PANTHER Ultrio Elite, Optitrol NAT HIV Screening DM21088 by reagent lot.

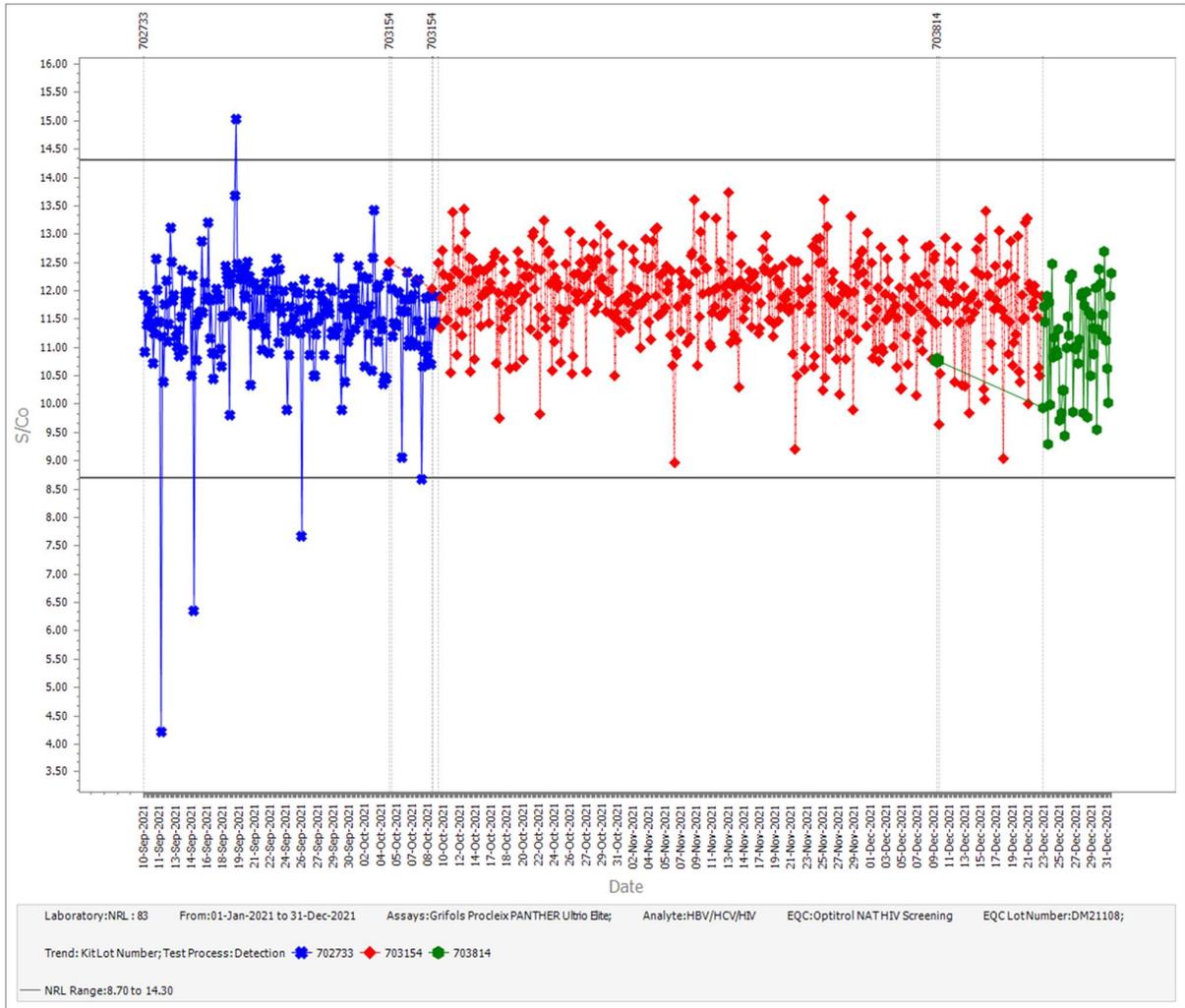


Figure 33 - Participant 83 data for Grifols Procleix PANTHER Ultrio Elite, Optitrol NAT HIV Screening DM21108 by reagent lot.

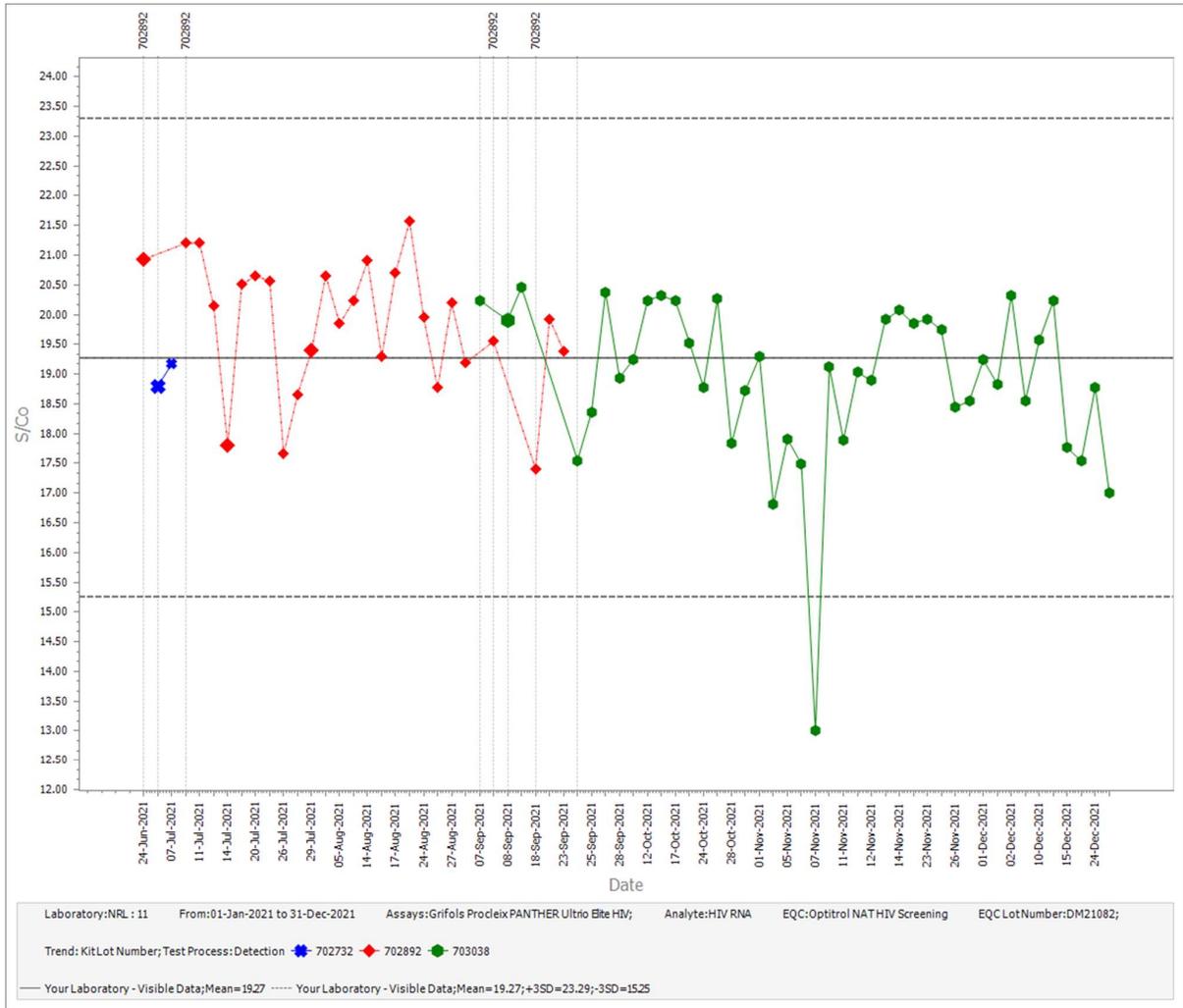


Figure 34 - Participant 11 data for Grifols Procleix PANTHER Ultrio Elite HIV, Optitrol NAT HIV Screening DM21082 by reagent lot.

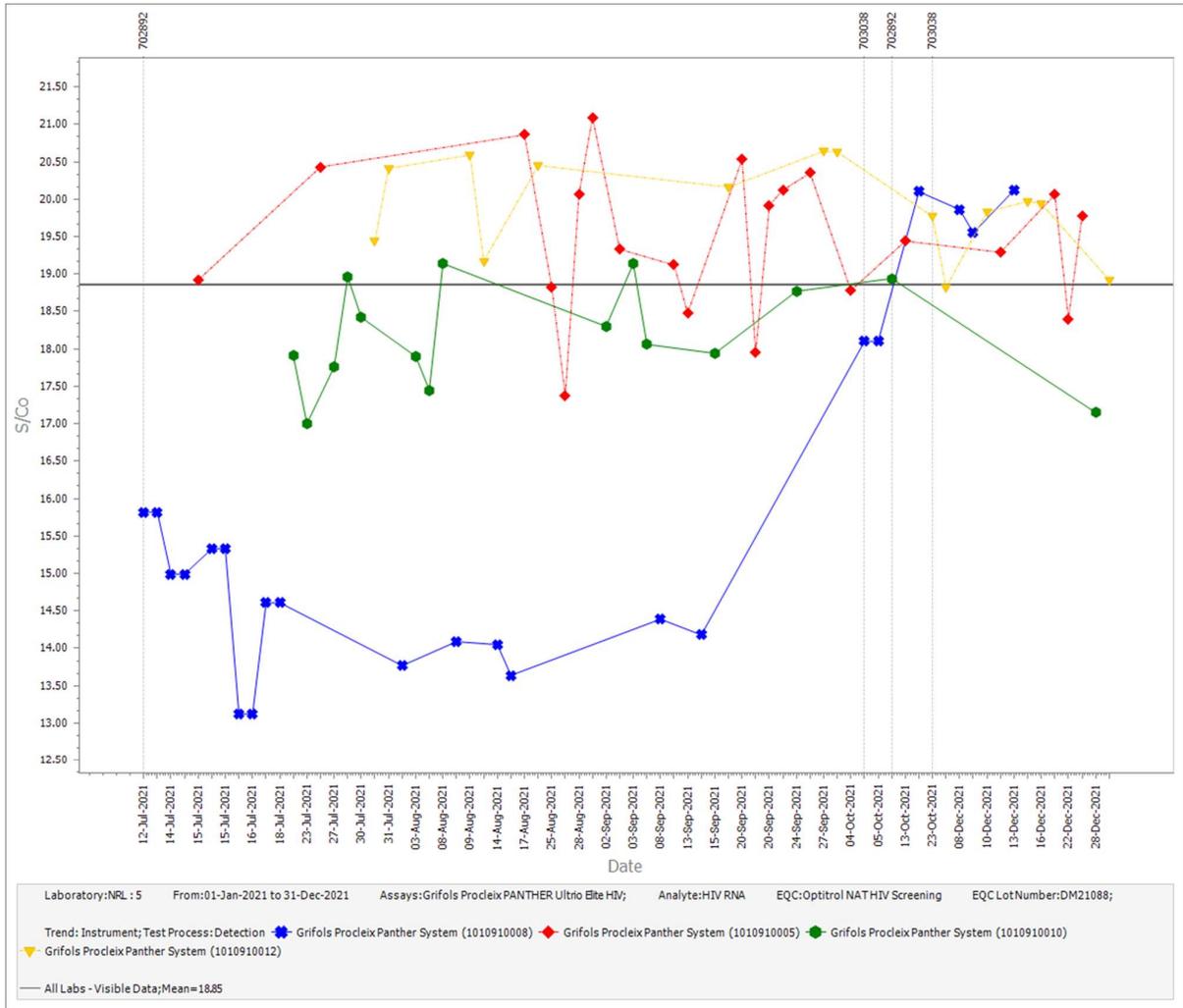


Figure 35 - Participant 5 data for Grifols Procleix PANTHER Ultrio Elite HIV, Optitrol NAT HIV Screening DM21088 by instrument.

EQC Lot	Total No. of Labs	Total	Drop	%
DM20017	10	1871	72	3.85%
DM20232	17	1999	341	17.06%
DM21082	4	1395	16	1.15%
DM21088	15	2771	33	1.19%
DM21108	12	2022	18	0.89%

Table 1: Summary of EQC lot number. Drops are results below 8.7 S/Co.

NRL Lab #	Reagent Lot						Grand Total		
	702892			703038					
	Total	Drop	%	Total	Drop	%	Total	Drop	%
1	142	8	5.63%	100	1	1.00%	242	9	3.72%
5	2	0	0.00%	194	5	2.58%	196	5	2.55%
Grand Total	144	8	5.56%	294	6	2.04%	438	14	3.20%

Table 2: AcroMetrix HIVRNA+ 221515 by lab and reagent lot number. Drops are results below 8.7 S/Co.

NRL Lab #	QC Lot									Grand Total		
	DM21082			DM21088			DM21108					
	Total	Drop	%									
1	437	3	0.69%							437	3	0.69%
3	157	2	1.27%	306	3	0.98%	1	0	0.00%	464	5	1.08%
5	81	1	1.23%	709	8	1.13%				790	9	1.14%
11	720	10	1.39%							720	10	1.39%
83							754	5	0.66%	754	5	0.66%
138				286	5	1.75%				286	5	1.75%
290				202	3	1.49%				202	3	1.49%
335				202	2	0.99%	389	3	0.77%	591	5	0.85%
359				78	1	1.28%	63	0	0.00%	141	1	0.71%
367				84	0	0.00%	54	3	5.56%	138	3	2.17%
371				52	0	0.00%	52	0	0.00%	104	0	0.00%
377				41	1	2.44%	88	0	0.00%	129	1	0.78%
378				112	4	3.57%				112	4	3.57%
380				98	0	0.00%	44	1	2.27%	142	1	0.70%
382				178	0	0.00%	389	4	1.03%	567	4	0.71%
385				110	0	0.00%				110	0	0.00%
446				165	6	3.64%	27	0	0.00%	192	6	3.13%
495				148	0	0.00%				148	0	0.00%
817							98	0	0.00%	98	0	0.00%
826							63	2	3.17%	63	2	3.17%
Grand Total	1395	16	1.15%	2771	33	1.19%	2022	18	0.89%	6188	67	1.08%

Table 3: Data breakdown by NRL lab and EQC lot number. Drops are results below 8.7 S/Co.

Reagent Lot	QC Lot									Grand Total		
	DM21082			DM21088			DM21108					
	Total	Drop	%									
702732	211	4	1.90%	4	0	0.00%	27	0	0.00%	242	4	1.65%
702733				61	0	0.00%	290	5	1.72%	351	5	1.42%
702892	792	6	0.76%	874	10	1.14%				1666	16	0.96%
702894				191	2	1.05%	4	0	0.00%	195	2	1.03%
703038	392	6	1.53%	191	1	0.52%				583	7	1.20%
703154				582	7	1.20%	824	3	0.36%	1406	10	0.71%
703461				122	0	0.00%	136	1	0.74%	258	1	0.39%
703564				409	5	1.22%	437	3	0.69%	846	8	0.95%
703814				301	6	1.99%	252	5	1.98%	553	11	1.99%
703890				13	1	7.69%	52	1	1.92%	65	2	3.08%
704272				10	0	0.00%				10	0	0.00%
704537				11	1	9.09%				11	1	9.09%
704564				2	0	0.00%				2	0	0.00%
Grand Total	1395	16	1.15%	2771	33	1.19%	2022	18	0.89%	6188	67	1.08%

Table 4: Data breakdown by reagent lot number and EQC lot number. Drops are results below 8.7 S/Co.