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**LONDON UNDERGROUND - ZOONO ANTIMICROBIAL TREATMENTS
TRIAL REPORT**

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Report Number: 02/15

17 September 2015

LONDON UNDERGROUND – ZOONO ANTIMICROBIAL TREATMENT TRIAL

Zoono UK Limited TRIAL REPORT No: 02/15

EXECUTIVE SUMMARY

Trial Author:	Mr James Milnes	Trial Dates:	27 Aug – 21 Sep 2015
Trial Report No:	02/15	Tel No:	

Introduction

1. London Underground Ltd requested Zoono UK Limited to conduct 'live' trials to provide accurate and tangible results in order to assess the effectiveness of Zoono as an antimicrobial treatment for potential use on the London Underground System. A trial plan was structured to assess performance and resilience of Zoono against a range of agreed tests.

Trial Aim.

2. The aim of the trial was to assess the performance the Zoono Antimicrobial capability against the current cleaning and sanitisation protocols. This would provide tangible results, which could be used to assess the applicability of the Zoono capability for protecting staff and customers utilising the London Underground.

3. **Objectives.** The trial objectives were to:

- a. Assess the performance of Zoono Antimicrobial Capability over a 21-day period.
- b. Make comments and recommendations based on the findings of the trial.

4. **Conclusion**

- a. The trial was a success and proved that the Zoono product remains active for more than the required 21-Day period.
- b. The carriages on the Underground are significantly contaminated and would need regular cleaning on an almost daily basis in order to achieve the same results as the Zoono product.

5. **Recommendations.** The Zoono capability was proven to be a success. A number of observations were made by the trials team and those comments are contained in the Main Report. A summary of observations are as follows:

- a. Zoono achieved 100% effectiveness over a 21-day period.
- b. 27.5% of carriage compartments are clean.
- c. 72.5% of carriage compartments are considered as contaminated.

LONDON UNDERGROUND – ZOONO ANTIMICROBIAL TREATMENT TRIAL

Zoono UK Limited TRIAL REPORT No: 02/15

MAIN REPORT

Trial Author: Mr James Milnes **Trial Dates:** 27 Aug – 21 Sep 2015
Trial Report No: 02/15 **Tel No:**

Background

1. Zoono Antimicrobial Treatments represent the next generation in surface and hand sanitisers providing protection for upto 28 days on surfaces and 24 hours on the skin. According to the National Security Risk Register the biggest threat to the United Kingdom is the threat posed by a long overdue Influenza Pandemic alongside the threat of Terrorist Attack. Coupled with the emerging threat of antibacterial resistant bacteria the importance of capabilities such a Zoono cannot be overstated enough. The unique way in which Zoono works ensures lasting protection whilst killing by lysis thereby mitigating the threat of bacteria becoming resistant to it.

2. **Operating Process.** There are three functional areas that the Zoono Capability could be employed:

- a. **Rolling Stock.** This describes the moving platforms that are used on the Underground System for the movement of passengers and staff.
- b. **Staff and Welfare.** This describes the Staff and Personnel environment.
- c. **Infrastructure.** This describes the infrastructure that supports the London Underground operations such as Stations, Offices, Ablutions and Rest Areas.

3. The equipment used in the trial was as follows:

- a. **Zoono Microbe Shield.** Zoono Microbe Shield is a food safe, water based surface sanitiser that bonds to the surface to kill 99.99% of all common pathogens. Zoono Microbe Shield is easy to apply, is environmentally friendly and is safe to use around people, plants and pets alike.

Zoono Microbe Shield has been tested in numerous, accredited laboratories around the world and proven to be highly effective against a variety of pathogens including E.coli, Staph, Pseudomonas, H1N1, Salmonella, Norovirus and even the Hospital Superbugs MRSA, VRE and C-Diff. In addition, it has been proven to be highly effective against yeast, fungi and mould spores including the black mould (Stachybotrys). Zoono Group has over 100 Test Results on file confirming the efficacy and durability of Zoono products. Zoono Microbe Shield is easy to apply via a trigger pack and one application will normally provide durable surface protection for between 14 and 30 days.



b. **London Underground Rolling Stock.** The train stock used on the Central Line are of 1992 stock vintage. The dimensions of the carriages are: Length 16.25m, Width 2.62m, Height 2.87m.



4. **Trial Staff.** The trials staffs were drawn from Zoono UK Limited Technical Team.

5. **Climatic Conditions.**

a. **London Central Line Depot.** The User Trial phase was conducted over 21 days from 27 August – 21 Sept 2015. Weather information was gathered from Zoono UK Limited Metrological equipment. Temperatures ranged from 14°C to 27°C during the day and at night highs of 14°C and lows of 6°C.

Limitations

6. **Environment.** The following limitations are associated with the trial environments:

a. There were no limitations to the trial environments.

7. **Locations.** The locations/touch points for the trail were chosen in order that a varied yet fair representation of the rolling stock was tested. Moreover, the locations identified were those most likely to be touched by both staff and travellers.

Swab Testing

8. EU Legislation states “Sampling of the environment can be a useful tool to identify and prevent the presence of pathogenic microorganisms”. The environment can be contaminated with a variety of micro-organisms derived from various sources. Estimation of the overall numbers of bacteria (Aerobic Colony Count) present can provide useful information when assessing general hygiene. Environmental monitoring can also be used as part of routine inspections of premises or in an investigation of a suspected viral outbreak where surfaces are thought to be likely vehicles of cross-contamination. Determination of the number of aerobic viable micro-organisms and the number of Enterobacteriaceae on a specified area of a surface can provide an indication of the cleanliness and allows monitoring of cleaning procedures over time.

9. The main value of determining the colony counts on a surface is to assess the cleanliness of that surface. A Detection and enumeration of bacteria in swab samples with a high count can be an early warning of inadequate cleaning practices.

10. Results:

Ser	CFU Count	Result
1	<10	Clean
2	10-160	Contaminated
3	>160	Gross Contamination

Trial Conduct

11. Scope and Method.

- a. **Objectives.** The trial objectives were to:
 - (1) Assess the performance of the Zoono Antimicrobial product over a minimum of a 21-day period. The 21-day period was chosen as this matched the current cleaning cycle of the carriages.
 - (2) Create a baseline of sample results in order to score the Zoono product against.
- b. **Scope.** The scope of the trial was to assess and test the Zoono Antimicrobial Product and provide user feedback and comment.
- c. **Method/Concept.** The method employed was to split the trial into two parts:
 - (1) **Baseline Tests.** This phase was conducted by Zoono UK Limited's trials team at the London Underground Central Line depot. Two 'double-cab' carriages were selected by the London underground staff. The team identified 'touch points'/sampling locations within each cab. It was agreed that sampling would take place before the regular cleaning process was undertaken and then again after the cleaning process and the application of Zoono to two of the carriages. This would provide an initial data set of 40 samples. The samples were then sent to an independent lab for analysis. Each of the touch points were swabbed and recorded in accordance with the data sheets.



Figure 1 – Touch Point Sampling

- (2) **21-Day Trial Point.** This phase was planned to be conducted at the 21-day point as the carriages were returned to the depot as part of cleaning cycle. However, due to circumstances outside of reasonable control the carriages could not be returned to the depot at the 21-day point. This portion of the trial was subsequently undertaken at the 25-day point. Each of the 40 identified sampling locations were sampled again and the samples sent to the independent laboratory for testing.



Figure 2 – Zoono UK Limited Trials Team Trials

d. **Measures of Effectiveness/Performance.** Data was collected, collated and assessed and then each data set was compared in order to see the effectiveness of Zoono. The results of which are shown at Annex A.

Results

12. The results section will be split into 3 separate sections to correspond with each Sample Set.

a. **Baseline Test – Pre Cleaning:**



Figure 3 – Application of Zoono

(1) **Objective and Test Methodology.** The plan was to test the carriages prior to cleaning in order to establish a baseline. Once tested Zoono was applied to the drivers cabs and two of the carriages. The agreed methodology is at Annex B.

(2) **Results.**

(a) The results showed that prior to the application of Zoono and after a 21-day's 'in service' and prior to cleaning 27.5% of the carriages were clean and 72.5% were contaminated Figure 4 (32.5% were contaminated and 40% were Grossly Contaminated – Figure 5).

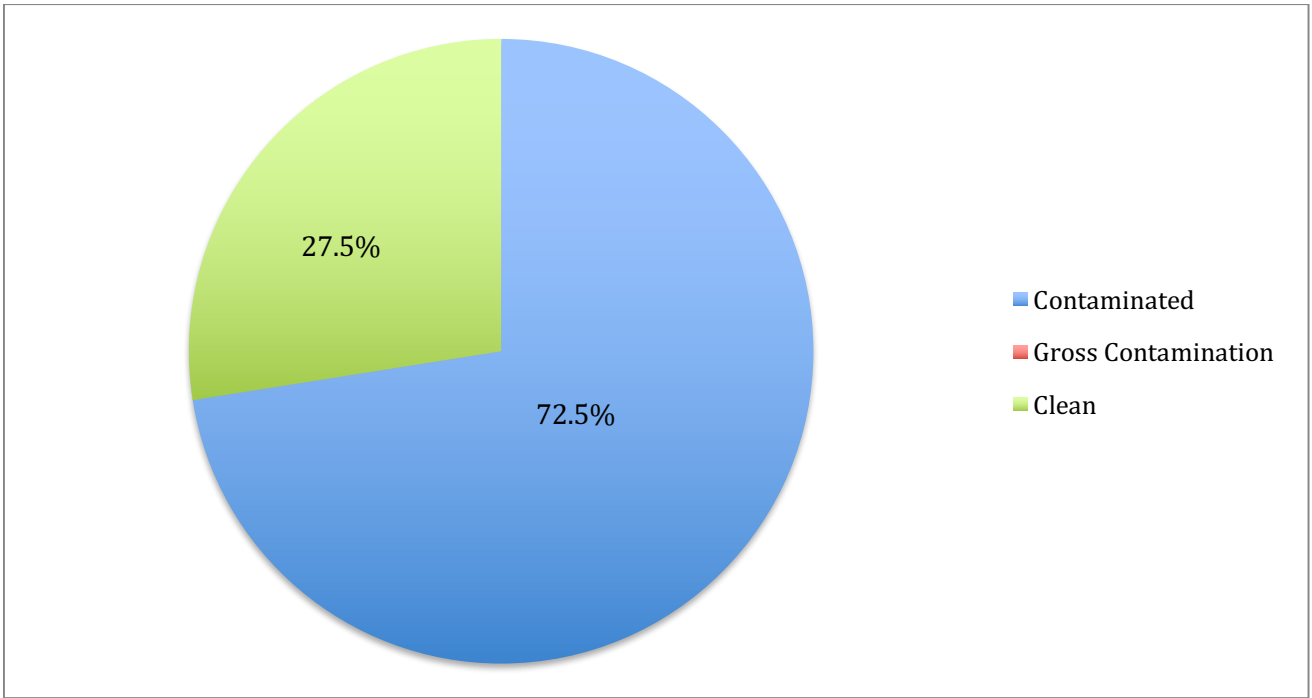


Figure 4 – 27 August - Overall Conatmination

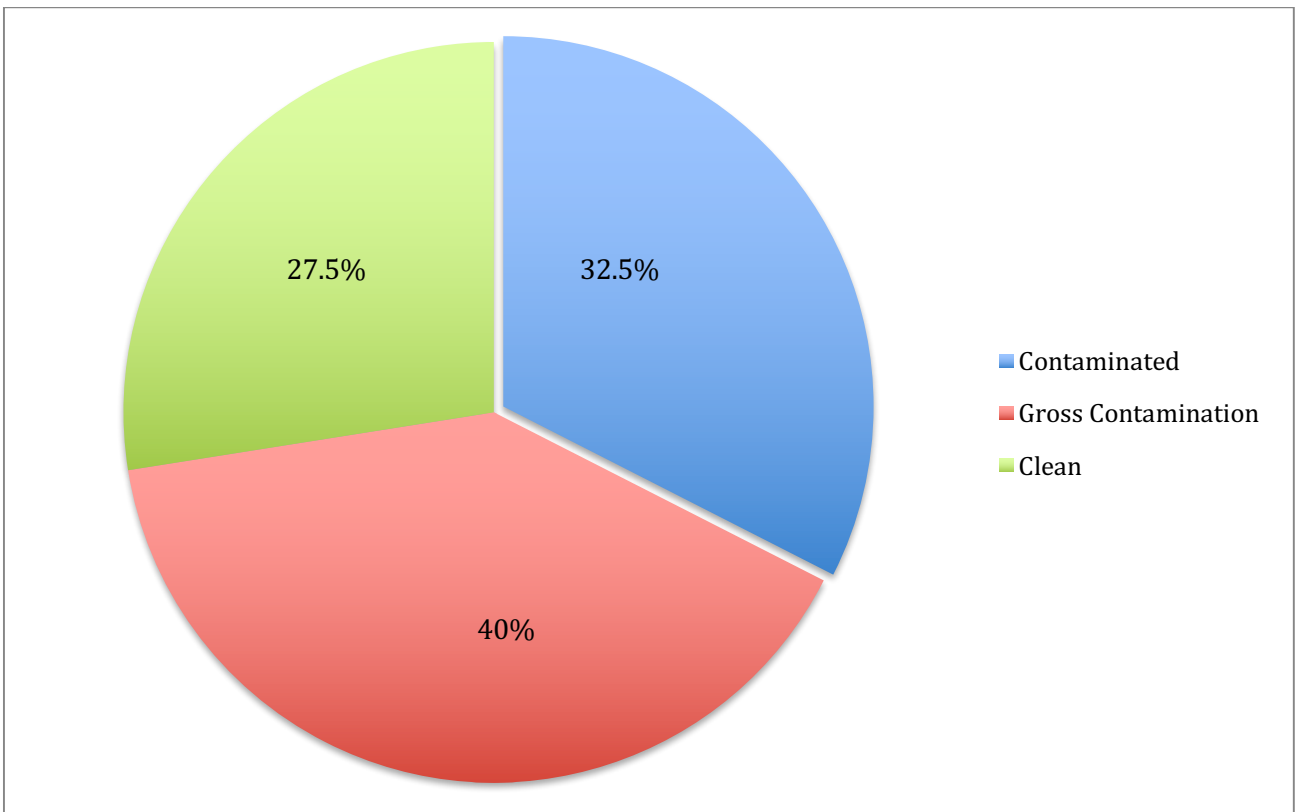


Figure 5 – 27 August – Contamination Breakdown

Comment.

(b) The surfaces swabbed are not considered as surfaces that would naturally harbour bacteria and therefore any positive results are mean that bacteria has been 'placed' on the surface by contact and cross contamination. Results by swab location - Figure 6 Overall Results, Figure 7 <10000 Results, Figure 8 <500 Results.

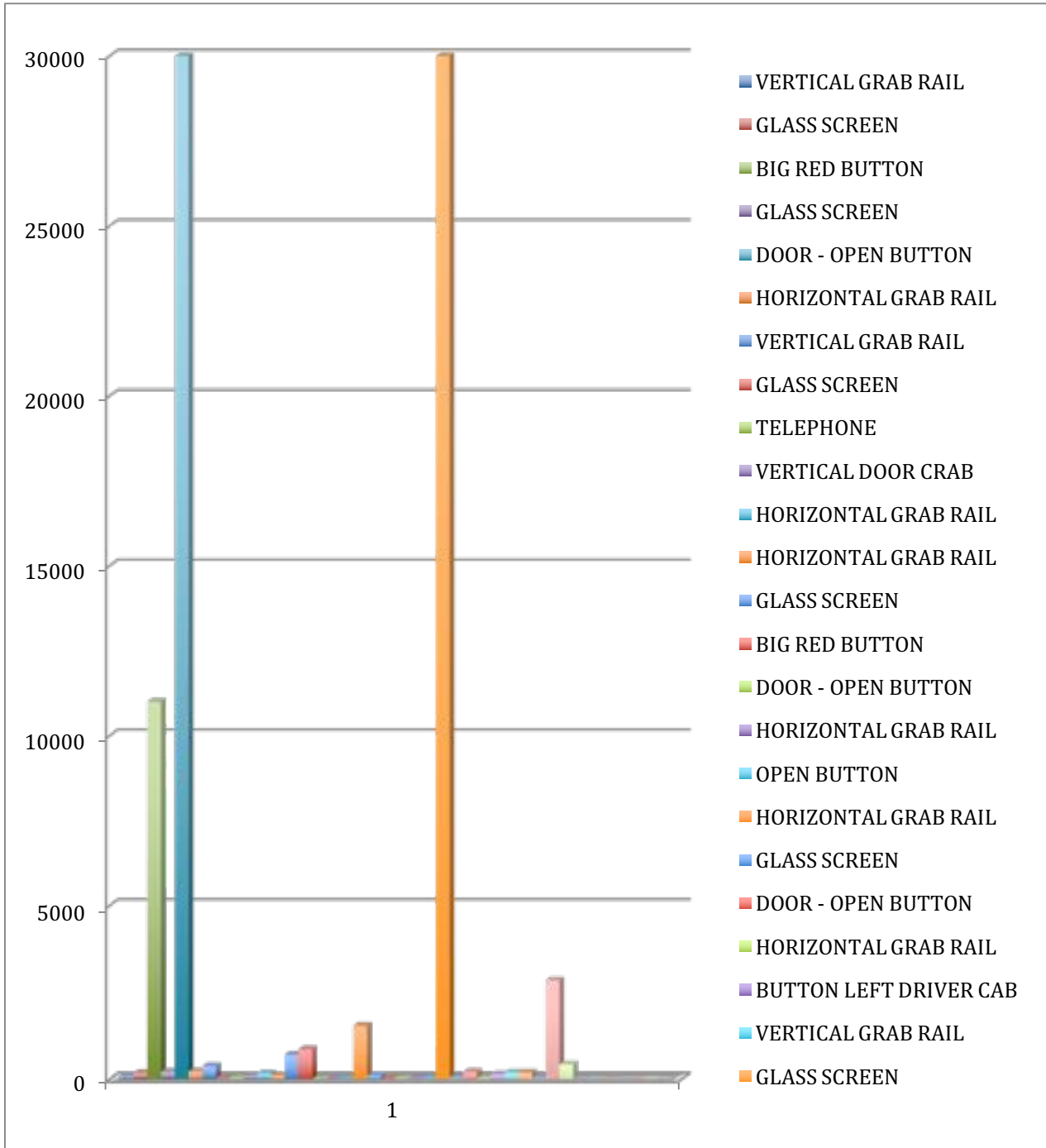


Figure 6 – Overall Results

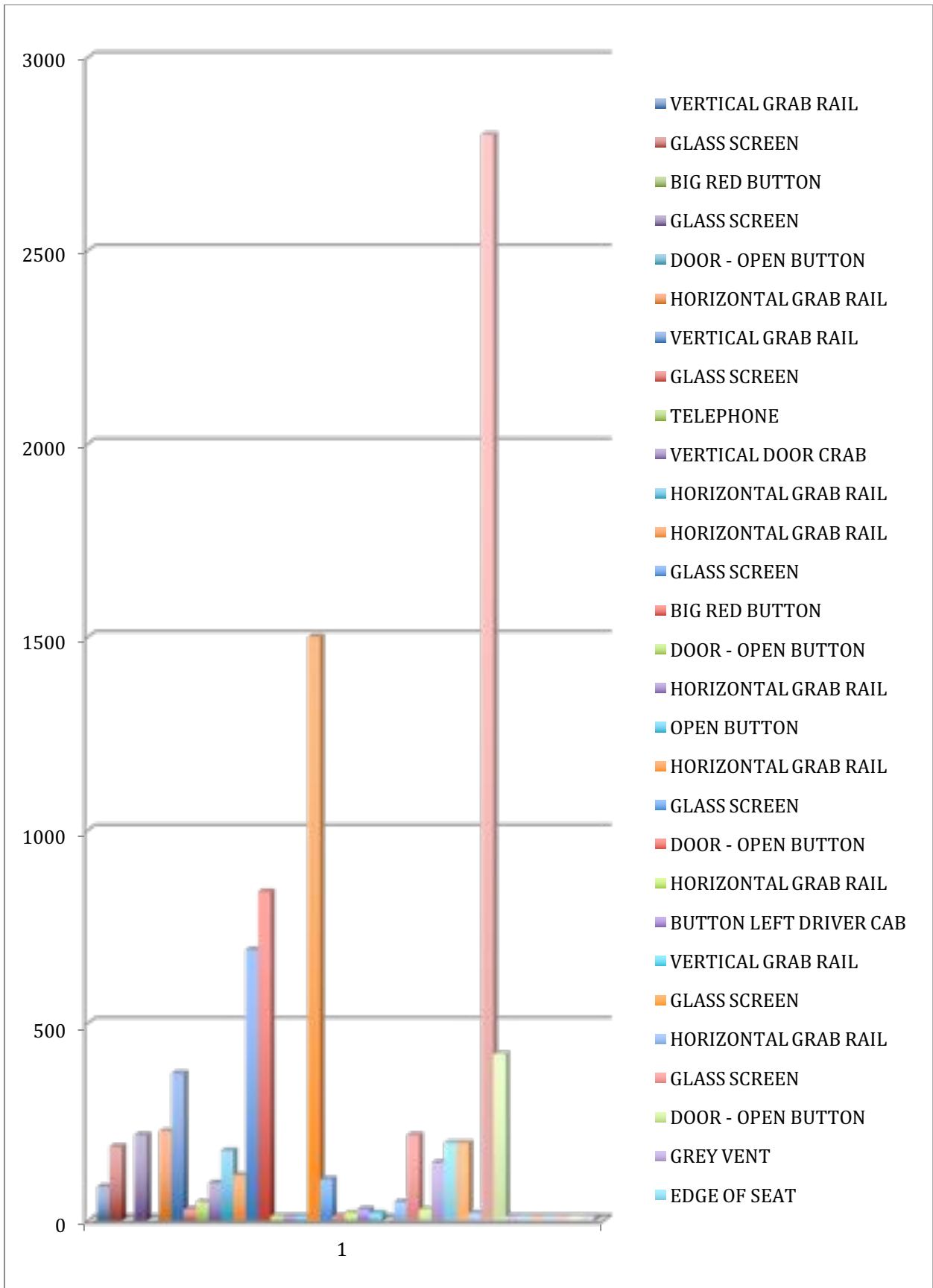


Figure 8 <10,000 Results

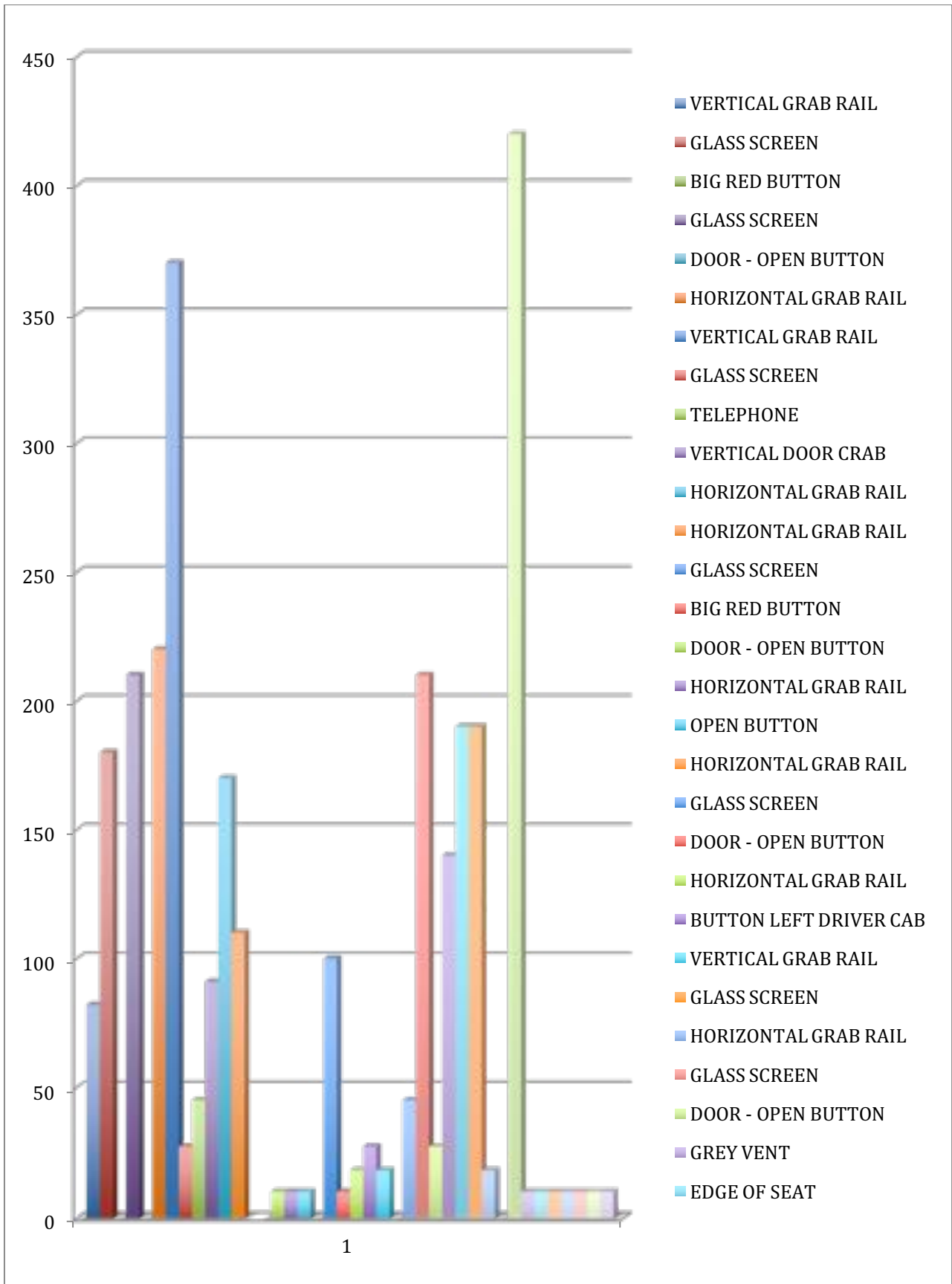


Figure 9 - <500 Results

Comment.

(c) Whilst there were nil results on the presence of Coagulase Positive Staphylococcus and Enterobacteriaceae the high results would suggest that there is a very high probability of other pathogenic bacteria being present.

b. **21 Day Tests:**

(1) **Objective and Test Methodology.** The plan was to take a set of 40 samples 20 from the Zoono carriages and 20 from the non-Zoono carriages. This would demonstrate the effectiveness or not of the Zoono product.

(2) **Results.**

(a) **Zoono.** The results showed that after the application of Zoono and the stack being in service for 25 days the Zoono swabbed areas indicated <10 score which is considered clean – Figure 10.

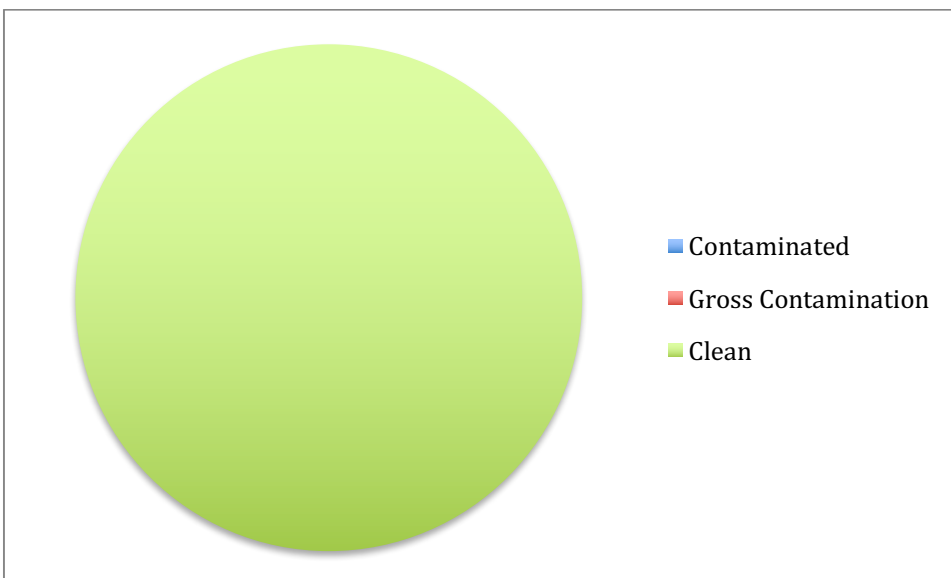


Figure 10 – Zoono Results

Comment. Zoono performed as expected.

(b) **Non-Zoono.** The results from the Non-Zoono swabs indicated that the carriages remained significantly contaminated with 35% being clean and 65% considered contaminated - Figure 11 Overall Results, (45% contamination 20% Gross Contamination) Figure 12 – Contamination Breakdown.

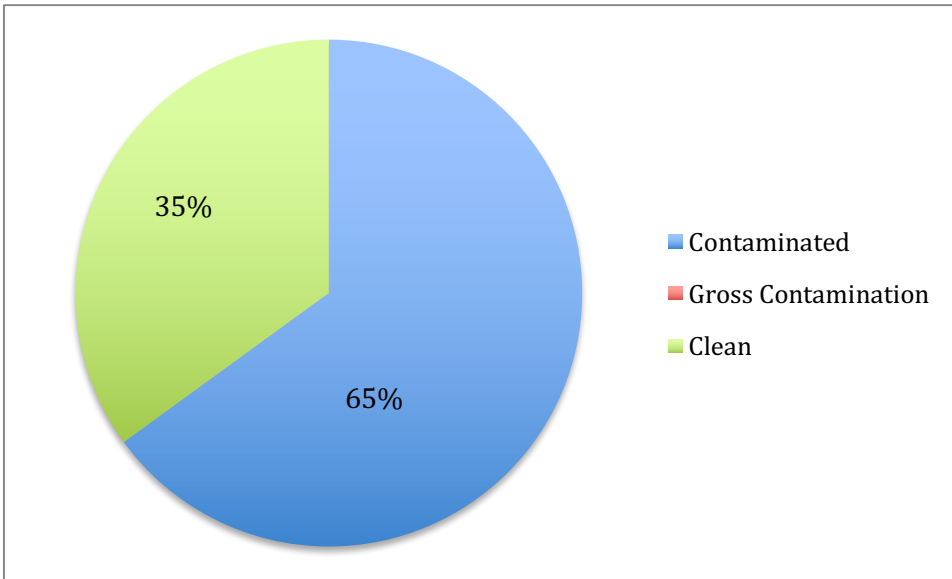


Figure 11 – Overall Contamination

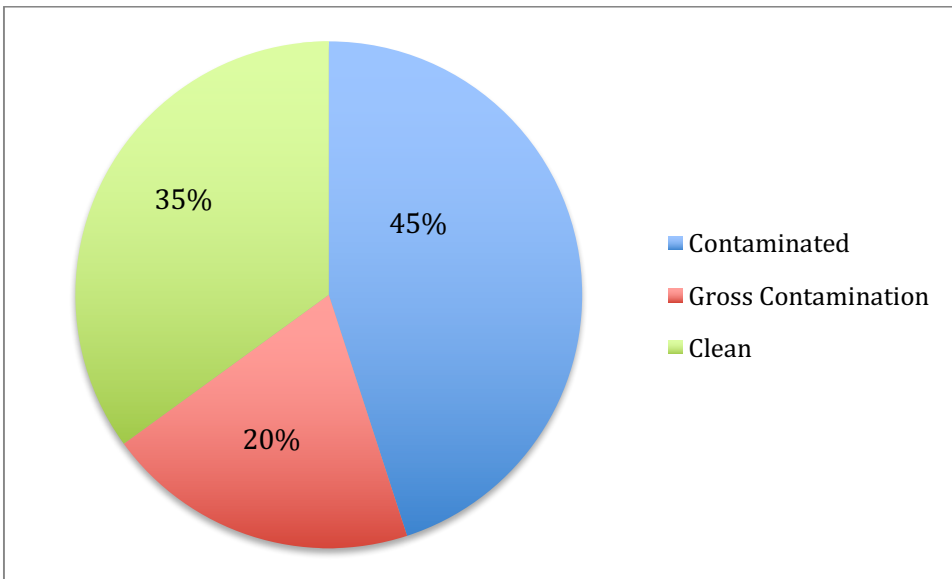


Figure 12 – Contamination Breakdown

Comment. The surfaces swabbed are not considered as surfaces that would naturally harbour bacteria and therefore any positive results are mean that bacteria has been 'placed' on the surface by contact and cross contamination. Results by swab location - Figure 13 Overall Results, Figure 14 <10000 Results, Figure 15 <600 Results.

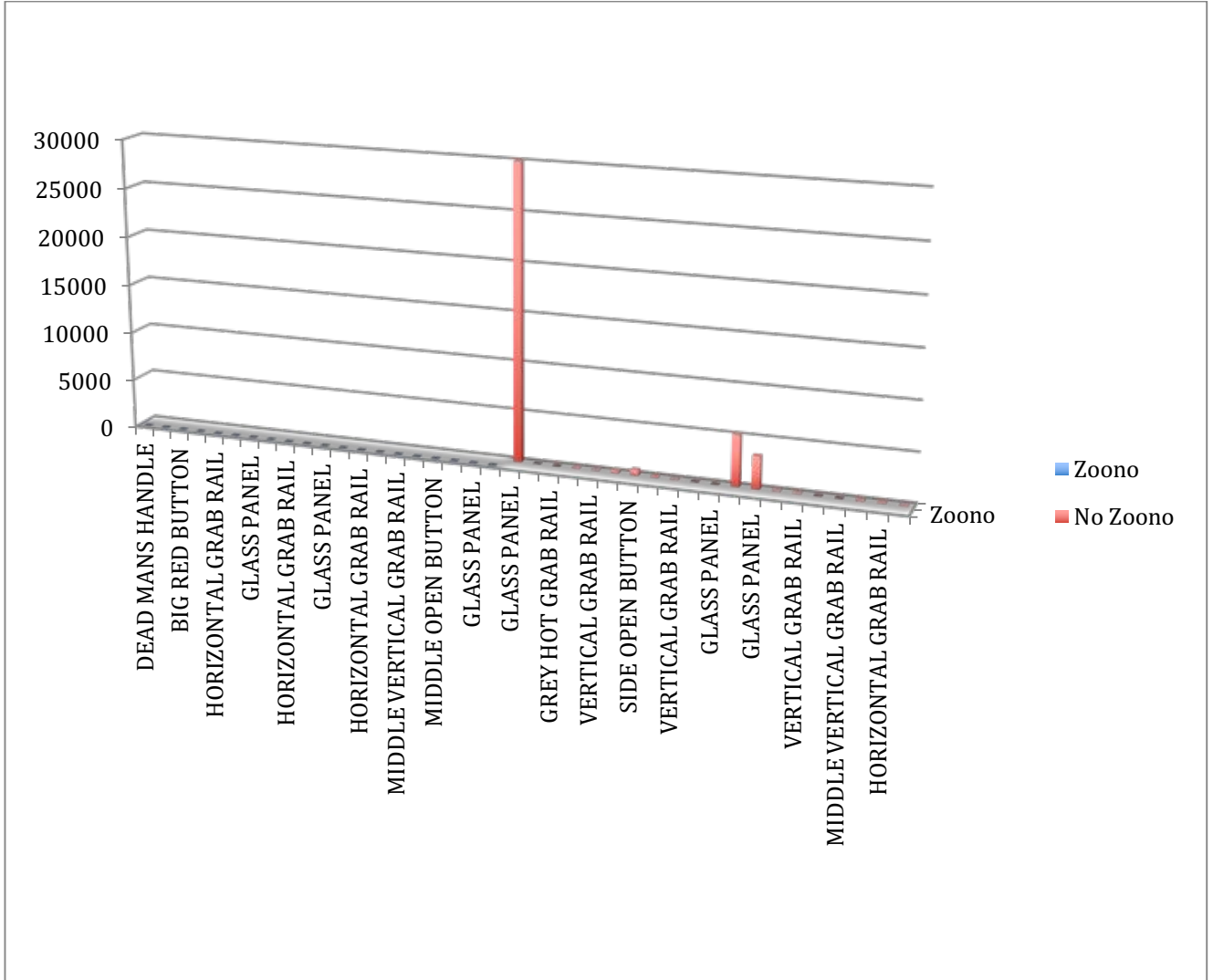


Figure 13 - Overall Results 21 September

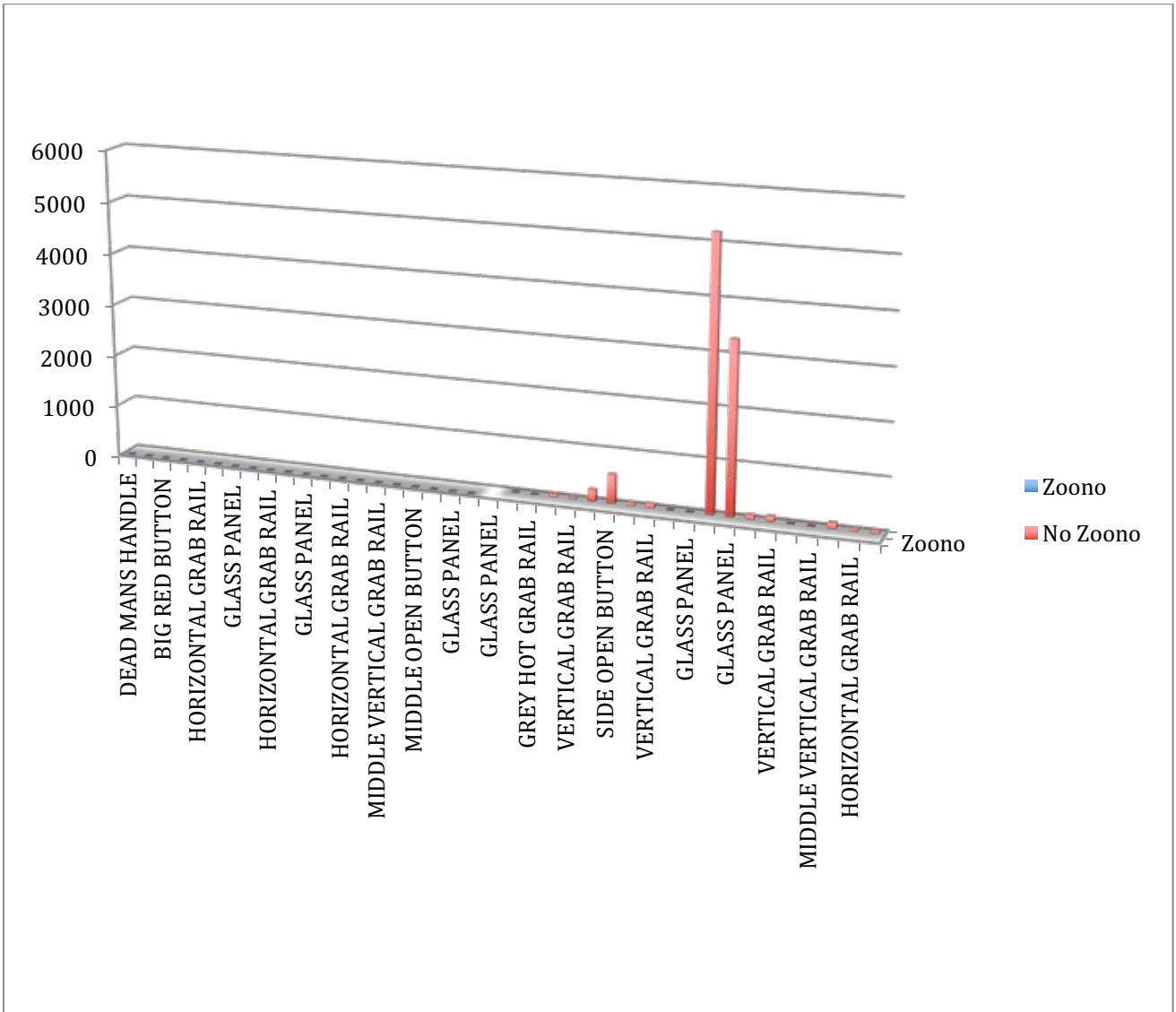


Figure 14 - <6000 Results

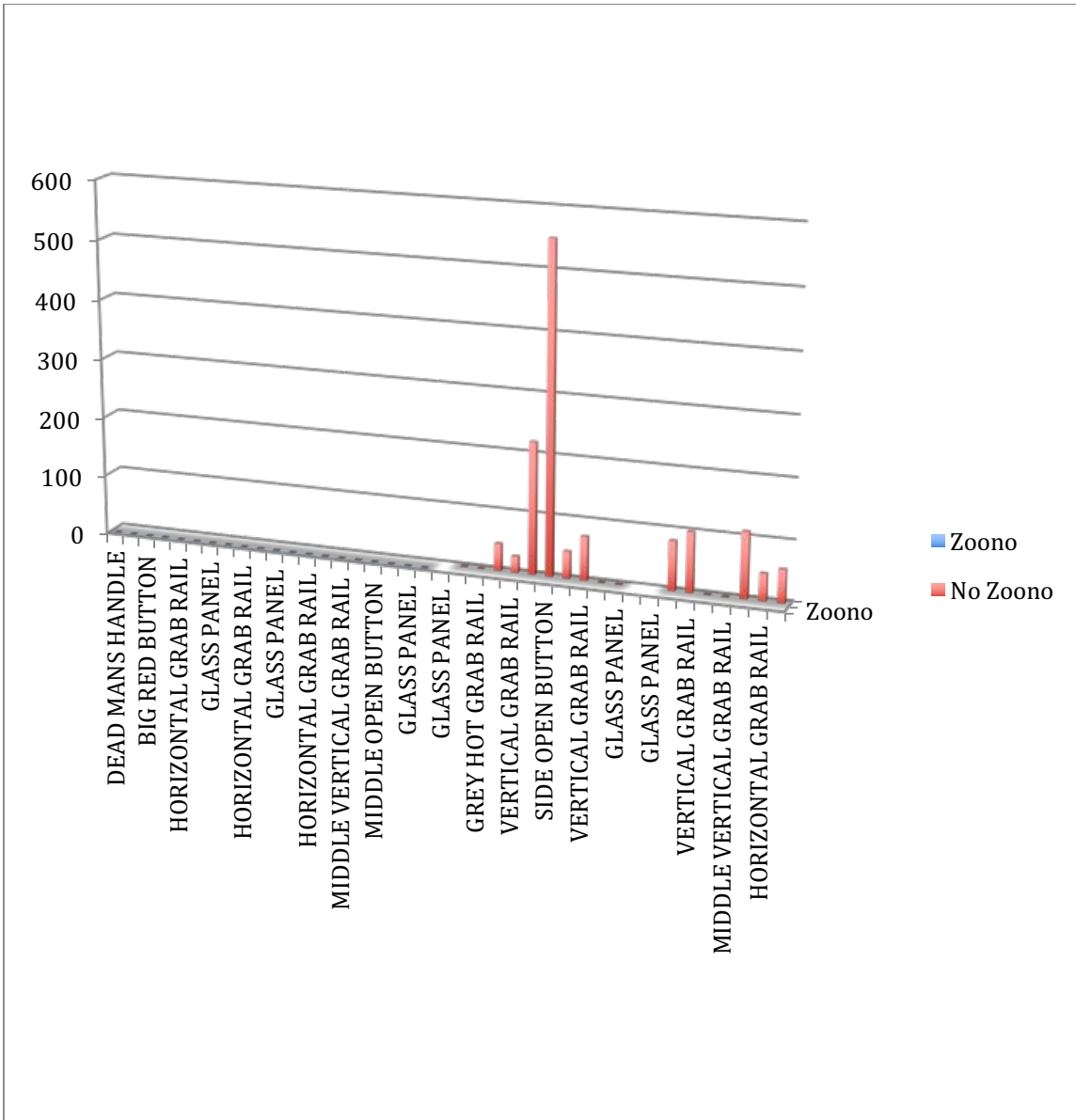


Figure 15 – <600 results

13. **Comments.** The following general comments were made about the performance of the Zoono product:

- a. Where Zoono was applied the results prove that Zoono remains an effective barrier against bacteria for over 21 days.
- b. The application of Zoono will significantly enhance the cleanliness of carriages and working environments.

14. **Conclusion**

- a. The trial was successful in testing fully the requirements detailed by the London Underground and ensured that areas of interest from the client were addressed, namely effectiveness of Zoono and duration of effectiveness.

b. In summary, the trial can recommend that Zoono would meet the user expectations and has exceeded the specified requirements. If brought into service, Zoono would provide the protection levels required on the London Underground for protecting staff and travellers.

15. **Recommendations.** It is recommended that further consideration is now given to the implementation of a Zoono cleaning regime on the London Underground Rolling Stock, Stations and support infrastructure.

DETAILED TEST RESULTS – 27 AUGUST 2015

Analytical Report Code	Lab. Sample Number	Client code	Client Reference	Aerobic Plate Count	Coagulase Positive Staphylococcus	Enterobacteriaceae
AR-15-UD-301084-01	400-2015-60236300	1.1.1	HORIZONTAL GRAB RAIL	1500	<10	<10
AR-15-UD-301090-01	400-2015-60236309	1.10.1	TELEPHONE	<10	<10	<10
AR-15-UD-301091-01	400-2015-60236310	1.11.1	BUTTON LEFT DRIVER CAB	27	<10	<10
AR-15-UD-302475-01	400-2015-60236311	1.12.1	BIG RED BUTTON	11000	<10	<10
AR-15-UD-302472-01	400-2015-60236301	1.2.1	VERTICAL GRAB RAIL	82	<10	<10
AR-15-UD-301085-01	400-2015-60236302	1.3.1	GLASS SCREEN	100	<10	<10
AR-15-UD-301086-01	400-2015-60236303	1.4.1	DOOR - OPEN BUTTON	<10	<10	<10
AR-15-UD-301087-01	400-2015-60236304	1.5.1	HORIZONTAL GRAB RAIL	18	<10	<10
AR-15-UD-301088-01	400-2015-60236305	1.6.1	VERTICAL GRAB RAIL	<10	<10	<10
AR-15-UD-302473-01	400-2015-60236306	1.7.1	GLASS SCREEN	180	<10	<10
AR-15-UD-302474-01	400-2015-60236307	1.8.1	DOOR - OPEN BUTTON	<10	<10	<10
AR-15-UD-301089-01	400-2015-60236308	1.9.1	DEAD MANS HANDLE	<10	<10	<10
AR-15-UD-302476-01	400-2015-60236312	2.1.1	HORIZONTAL GRAB RAIL	<10	<10	<10
AR-15-UD-301092-01	400-2015-60236313	2.2.1	VERTICAL GRAB RAIL	18	<10	<10
AR-15-UD-302477-01	400-2015-60236314	2.3.1	GLASS SCREEN	210	<10	<10
AR-15-UD-302478-01	400-2015-60236315	2.4.1	DOOR - OPEN BUTTON	>30000	<10	<10
AR-15-UD-302479-01	400-2015-60236316	2.5.1	HORIZONTAL GRAB RAIL	220	<10	<10
AR-15-UD-302480-01	400-2015-60236317	2.6.1	VERTICAL GRAB RAIL	370	<10	<10
AR-15-UD-301093-01	400-2015-60236318	2.7.1	GLASS SCREEN	>30000	<10	<10
AR-15-UD-301094-01	400-2015-60236319	2.8.1	DOOR - OPEN BUTTON	<10	<10	<10
AR-15-UD-302482-01	400-2015-60236329	3.10.1	TELEPHONE	45	<10	<10
AR-15-UD-301095-01	400-2015-60236320	3.1.1	HORIZONTAL GRAB RAIL	45	<10	<10
AR-15-UD-302540-01	400-2015-60236330	3.11.1	BIG RED BUTTON	840	<10	<10
AR-15-UD-301103-01	400-2015-60236331	3.12.1	BUTTON CLOSE SALOON	<10	<10	<10
AR-15-UD-301096-01	400-2015-60236321	3.2.1	VERTICAL GRAB RAIL	<10	<10	<10
AR-15-UD-301097-01	400-2015-60236322	3.3.1	GLASS SCREEN	210	<10	<10
AR-15-UD-301098-01	400-2015-60236323	3.4.1	DOOR - OPEN BUTTON	27	<10	<10
AR-15-UD-301099-01	400-2015-60236324	3.5.1	GREY VENT	140	<10	<10
AR-15-UD-301100-01	400-2015-60236325	3.6.1	EDGE OF SEAT	190	<10	<10
AR-15-UD-302481-01	400-2015-60236326	3.7.1	GLASS SCREEN	27	<10	<10
AR-15-UD-301101-01	400-2015-60236327	3.8.1	OPEN DOOR BUTTON	190	<10	<10
AR-15-UD-301102-01	400-2015-60236328	3.9.1	DEAD MANS HANDLE	18	<10	<10
AR-15-UD-302483-01	400-2015-60236332	4.1.1	VERTICAL DOOR GRAB	91	<10	<10
AR-15-UD-302484-01	400-2015-60236333	4.2.1	HORIZONTAL GRAB RAIL	170	<10	<10
AR-15-UD-301104-01	400-2015-60236334	4.3.1	GLASS SCREEN	<10	<10	<10
AR-15-UD-301105-01	400-2015-60236335	4.4.1	OPEN BUTTON	2800	<10	<10
AR-15-UD-302485-01	400-2015-60236336	4.5.1	HORIZONTAL GRAB RAIL	110	<10	<10
AR-15-UD-301106-01	400-2015-60236337	4.6.1	VERTICAL GRAB RAIL	420	<10	<10
AR-15-UD-302486-01	400-2015-60236338	4.7.1	GLASS SCREEN	690	<10	<10
AR-15-UD-302487-01	400-2015-60236339	4.8.1	OPEN BUTTON	<10	<10	<10

DETAILED TEST RESULTS – NON-ZOONO – 21 SEPTEMBER 2015

Analytical Report Code	Lab. Sample Number	Client code	Client Reference	Aerobic Plate Count	Coagulase Positive Staphylococcus	Enterobacteriaceae
PR-15-UD-001899-01	400-2015-60257183	01	GLASS PANEL	30000	<10	<10
PR-15-UD-001900-01	400-2015-60257184	02	MIDDLE VERTICAL GRAB RAIL	<10	<10	<10
PR-15-UD-001901-01	400-2015-60257185	03	GREY HOT GRAB RAIL	<10	<10	<10
PR-15-UD-001902-01	400-2015-60257186	04	MIDDLE OPEN BUTTON	45	<10	<10
PR-15-UD-001903-01	400-2015-60257187	05	VERTICAL GRAB RAIL	27	<10	<10
PR-15-UD-001904-01	400-2015-60257188	06	HORIZONTAL GRAB RAIL	220	<10	<10
PR-15-UD-001905-01	400-2015-60257189	07	SIDE OPEN BUTTON	550	<10	<10
PR-15-UD-001906-01	400-2015-60257190	08	HORIZONTAL GRAB RAIL	45	<10	<10
PR-15-UD-001907-01	400-2015-60257191	09	VERTICAL GRAB RAIL	73	<10	<10
PR-15-UD-001908-01	400-2015-60257192	10	VERTICAL GRAB RAIL	<10	<10	<10
PR-15-UD-001909-01	400-2015-60257193	11	GLASS PANEL	<10	<10	<10
PR-15-UD-001910-01	400-2015-60257194	12	VERTICAL GRAB RAIL	5200	<10	<10
PR-15-UD-001911-01	400-2015-60257195	13	GLASS PANEL	3300	<10	<10
PR-15-UD-001912-01	400-2015-60257196	14	HORIZONTAL GRAB RAIL	82	<10	<10
PR-15-UD-001913-01	400-2015-60257197	15	VERTICAL GRAB RAIL	100	<10	<10
PR-15-UD-001914-01	400-2015-60257198	16	GLASS PANEL	<10	<10	<10
PR-15-UD-001915-01	400-2015-60257199	17	MIDDLE VERTICAL GRAB RAIL	<10	<10	<10
PR-15-UD-001916-01	400-2015-60257200	18	VERTICAL GRAB RAIL	110	<10	<10
PR-15-UD-001917-01	400-2015-60257201	19	HORIZONTAL GRAB RAIL	45	<10	<10
PR-15-UD-001918-01	400-2015-60257202	20	GREY HORIZONTAL GRAB RAIL	55	<10	<10

DETAILED TEST RESULTS – ZOONO TREATED – 21 SEPTEMBER 2015

Analytical Report Code	Lab. Sample Number	Client code	Client Reference	Aerobic Plate Count	Coagulase Positive Staphylococcus	Enterobacteriaceae
PR-15-UD-001919-01	400-2015-60257203	Z1	DEAD MANS HANDLE	<10	<10	<10
PR-15-UD-001920-01	400-2015-60257204	Z2	DRIVER SIDE BUTTON	<10	<10	<10
PR-15-UD-001921-01	400-2015-60257205	Z3	BIG RED BUTTON	<10	<10	<10
PR-15-UD-001922-01	400-2015-60257206	Z4	TELEPHONE	<10	<10	<10
PR-15-UD-001923-01	400-2015-60257207	Z5	HORIZONTAL GRAB RAIL	<10	<10	<10
PR-15-UD-001924-01	400-2015-60257208	Z6	VERTICAL GRAB RAIL	<10	<10	<10
PR-15-UD-001925-01	400-2015-60257209	Z7	GLASS PANEL	<10	<10	<10
PR-15-UD-001926-01	400-2015-60257210	Z8	OPEN BUTTON	<10	<10	<10
PR-15-UD-001927-01	400-2015-60257211	Z9	HORIZONTAL GRAB RAIL	<10	<10	<10
PR-15-UD-001928-01	400-2015-60257212	Z10	MIDDLE OPEN BUTTON	<10	<10	<10
PR-15-UD-001929-01	400-2015-60257213	Z11	GLASS PANEL	<10	<10	<10
PR-15-UD-001930-01	400-2015-60257214	Z12	MIDDLE VERTICAL GRAB RAIL	<10	<10	<10
PR-15-UD-001931-01	400-2015-60257215	Z13	HORIZONTAL GRAB RAIL	<10	<10	<10
PR-15-UD-001932-01	400-2015-60257216	Z14	VERTICAL GRAB RAIL	<10	<10	<10
PR-15-UD-001933-01	400-2015-60257217	Z15	MIDDLE VERTICAL GRAB RAIL	<10	<10	<10
PR-15-UD-001934-01	400-2015-60257218	Z16	GLASS PANEL	<10	<10	<10
PR-15-UD-001935-01	400-2015-60257219	Z17	MIDDLE OPEN BUTTON	<10	<10	<10
PR-15-UD-001936-01	400-2015-60257220	Z18	VERTICAL GRAB RAIL	<10	<10	<10
PR-15-UD-001937-01	400-2015-60257221	Z19	GLASS PANEL	<10	<10	<10
PR-15-UD-001938-01	400-2015-60257222	Z20	VERTICAL GRAB RAIL	<10	<10	<10