Sabretech QS (Cas no: 27668-52-6) (Armatrex, Penetrexx, Biotrexx 247, Prolyse 247)

# **SMART** With Sabretech QS, also known as; Armatrex, Penetrexx, Biotrexx 247, Prolyse 247 from Clearstream Technologies - Summary of Supportive Data

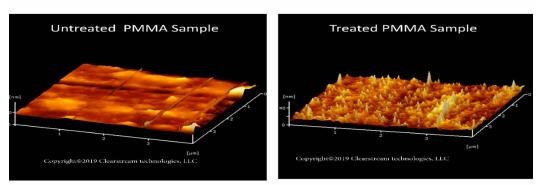
# **Treatment Delivery Methods and Treated Substrate Observations**

## Armatrex and Clearstream Alternative Brand Named Products

- The Clearstream suite of aligned branded products have been extensively and successfully applicated on multiple substrates without deformities, color loss, or surface material degradation.
- The Clearstream suite of aligned branded products have been tested and delivered successfully utilizing a myriad of methods such as dipping, wiping, brushing, and padding. Armatrex has also successfully applied with devices ranging from foggers, low pressure sprayers, high pressure sprayers, backpack coarse sprayers, trigger sprayers, and most importantly, electrostatic sprayers without clogging.
- The Clearstream suite of aligned branded products have been repeatedly and successfully tested for uniformity and polymerization of its coating, bonding characteristics, and durability. (See: South Carolina Hatchery (Hatcher) Durability Field Study below in Field Studies.)

# Atomic Force Microscopy (AFM), Light Microscopy, and Scanning Electron Microscopy (SEM)

#### AFM/PMMA Sample



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The above image on the left is a sample of an untreated **PMMA (Poly methyl methacrylate)** sample and the same **PMMA** sample (right image) treated with Armatrex (Clearstream's Sabretech QS). The treated sample was treated 24 hours before being viewed microscopically at 32,000x via an **Atomic Force Microscope** (AFM), and cured in a low temp oven by Dr. Lister.

- While AFM doesn't give us the ability to perform Elemental Detection, it does allow us to see the new topographic landscape. It will provide time lapse imagery and the sequence of events of untreated, treated, inoculation (time zero), and cell destruction at various intervals such as 10 minutes, 1 hour, 8 hours, or 24 hours post inoculation. In combination with 3<sup>rd</sup> party microbiology testing labs that do cell counts, this is an invaluable tool Clearstream will be acquiring.
- The sample area is 4 micrometers by 4 micrometers
- The average height of the molecule's carbon chains is approximately 5 nano-meters.
- In addition to the molecular spikes that we see in the topographic image, the darker colors represent the deeper topographic "valleys".
- Offending microorganisms whether lysed by the carbon chain or not, are drawn in by the cationic charge density that begins with the proton rich environment surrounding the 18 carbon chain.
- Once the microorganism descends into the valleys, the cytoplasmic destruction of the cell's membrane is completed. The cell is overwhelmed and exhausted by the polymerized nitrogen atoms that provide unavoidable destructive force.

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### Scanning Electron Microscopy (SEM)/PMMA Sample

Image 1

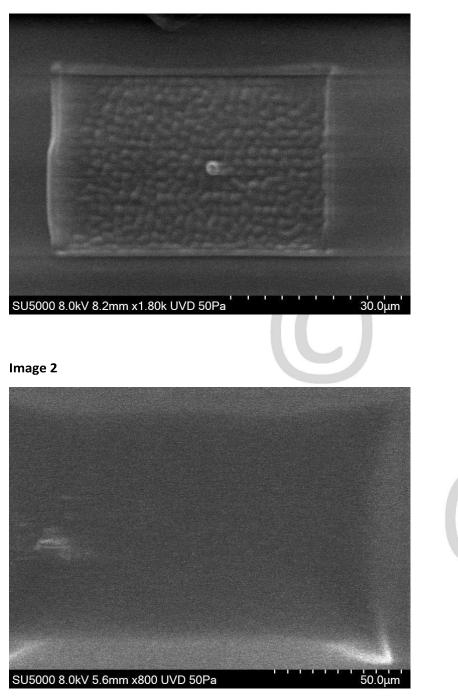


Image 1 and Image 2 are both one half of a Polymethyl methacrylate (PMMA) sample:

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- Image 1 is the untreated portion of the PMMA sample (control). It is one half of the sample coupon itself. You will see a corn row or cobble stone type of effect resulting from the electron beam moving back and forth in its scanning process of the surface. This effect is due to heat buildup and the non-reflective nature of the substrate which is described as "charging".
- This effect occurs because the electrons are trapped within the substrate causing it to roil up or basically melt from the inside out. This is caused by an accumulation of energy in the material itself due to absorption of the energy instead of deflecting the energy like a stainless steel sample might provide.
- Image 2 is the treated portion of the PMMA sample. Again, all of the specifications and settings for power and magnification were identical to the control scan. You will see an unmistakable difference in the scanned areas. The area scanned is a little larger than the scanned area of the control, but the effect is undeniable.
  - The scanned area developed a "pushed in" or concave pillow effect (think Styrofoam meat tray), but did not display the roiled or melted effect observed in the control sample. The Hitachi reps were very excited. They said while we weren't picking up the elemental detection we were looking for, we discovered something of valuable importance... it's clear that the coating provided a physical protective layer on the PMMA substrate.
  - While the electron beam clearly warped the substrate, the Armatrex SiQAC film did not separate from the PMMA surface, the polymer did not separate or break apart from itself, and it clearly provided a form of protection for the substrate. It appears that the SiQAC film diffused or deflected the electron beam sufficiently enough to avoid the melting effect that occurred on the untreated PMMA control half of the sample.
  - We have seen similar evidence in terms of the molecule's ability to survive under direct UV exposure. While the electron beam and UV rays are different from one another it is undeniable that the surface of the treated sample held up significantly better than the untreated control.
  - We are intrigued by this discovery and we are delving deeper into the components of the molecule and its polymerization to identify the

basis for the protective nature, build upon its obvious characteristics, figure out new commercial market applications, add to the technical exceptionalism of the technology, and advance it as best we can.

#### Note:

The following Images are taken from a study conducted by Dr. Jeanna Wilson in 2019 at the University of Georgia Poultry Science Department under the direction of Dr. Ivan Alvarado of MAH. Clearstream provided the Armatrex antimicrobial product and conducted the electrostatic application treatments. Specific goals and findings of the study are summarized further below in this document under the heading of Poultry Laboratory Testing.

#### Light Microscopy/Egg Shell Sample

Image 1. Light microscopy of a pore from an eggshell fragment from the control group.



Image 2. Light microscopy of a pore from an eggshell fragment treated with the 0.1% solution.



Image 3. Light microscopy of a pore from an eggshell fragment treated with the 0.25% solution.

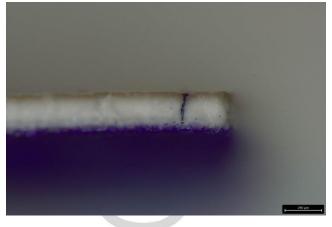


Image 4. Light microscopy of a pore from an eggshell fragment treated with the 0.5% solution.

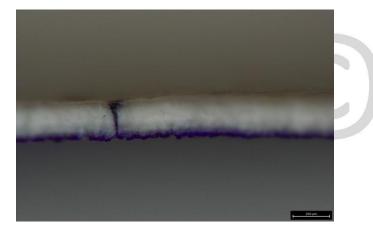
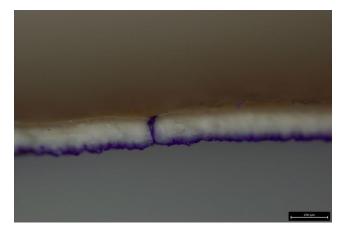


Image 5. Light microscopy of a pore from an eggshell fragment treated with the 1.0% solution.

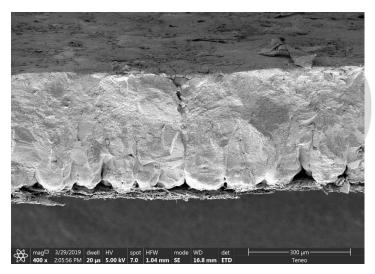


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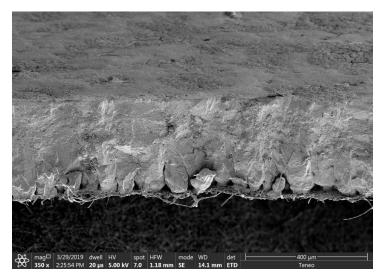
Evaluation of eggshell fragments from each treatment group resulted in pores that look similar across the treatments with no visible obstructions. However, results from this test were inconclusive because of the preparation method. Bleach (0.268 M Na hypochlorite) was used to dissolve the inner membrane of the eggshell, and in doing so could have made an impact on the pores. Because of this, it was necessary to do scanning electron microscopy on a new subset of eggshell fragments.

## SEM/Egg Shell Samples

**Image 6.** Image shows scanning electron microscopy (SEM) of an eggshell cross section from the control treatment. The top surface is the outermost portion of the eggshell and the bottom layer is the inner eggshell membrane.



**Image 7.** Image shows SEM of an eggshell cross section from the highest level treatment, 1.0%. The top surface is the outermost portion of the eggshell and the bottom layer is the inner eggshell membrane.



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The eggshell fragments from the control and highest level treatment groups had similar appearance when analyzed using SEM. The eggs appeared to have obvious crystalline patterns, indicating the palisade layer and mammillary knobs of the eggshell structure (Dombre et al., 2016). There was no obvious cuticle layer in most samples, nor an obvious layer of residue from the treatment application.

# XPS Microscopy/Upcoming Lab Sessions

Clearstream has entered into an agreement with Clemson University To utilize the university's extensive Electron Microscopy Laboratories including their XPS lab. Planned dates had to be canceled due to the current COVID 19 pandemic and the shutdown of most every higher learning institutions in the US. As soon as things begin to normalize we will reschedule the sessions and forward the data as we collect it.

Through our research and after trial and error, it appears that X-ray photoelectron spectroscopy (XPS), also known as electron spectroscopy for chemical analysis (ESCA), is the proper technique for analyzing the surface chemistry of a material. XPS can measure the elemental composition, empirical formula, chemical state and electronic state of the elements within a material.

In addition to AFM, Clearstream wishes to further identify the chemical compound of the Armatrex molecule once bound to various common substrates found in targeted equipment and structure treatments. Plastics, painted surfaces, composites, and stainless steel to name a few. Clearstream believes this type of imagery along with elemental detection graphics will enhance marketing efforts combined with documented performance.

# Laboratory Testing

# Microbiological Laboratory Challenge Testing

- The Clearstream suite of aligned branded products were tested at the Microchem Laboratory of Round Rock, Texas and proven effective against *Escherichia coli (E. coli)* at multiple contact times utilizing the standardized JIS Z 2801 test.
- The Clearstream suite of aligned branded products were tested at the Microchem Laboratory of Round Rock, Texas and proven effective against

Salmonella enterica at multiple contact times utilizing the standardized JISZ 2801 test.

- The Clearstream suite of aligned branded products were tested at the Microchem Laboratory of Round Rock, Texas and proven effective against *Campylobacter juni* at multiple contact times utilizing the standardized JISZ 2801 test.
- The Clearstream suite of aligned branded products were tested at the Microchem Laboratory of Round Rock, Texas and proven effective against *Candida albicans* at multiple contact times utilizing the standardized JISZ 2801 test.
- The Clearstream suite of aligned branded products were tested at the Microchem Laboratory of Round Rock, Texas and proven effective against *Staphylococcus aureus* in a specialized test with significant reductions at a 24 hour contact time incorporated into a semi porous conveyor belt material.
- The Clearstream suite of aligned branded products were tested at the former Antimicrobial Test Laboratory now known as the Microchem Laboratory of Round Rock, Texas and proven effective against *Staphylococcus epidermidis* at multiple contact times utilizing the standardized AATCC 100 test.

# Virus Testing

- Clearstream is currently working with Microchem Laboratories on three test variants of *Human Coronavirus 229E* against the Clearstream suite of aligned branded products. These are modifications of existing standardized bacterial tests for immobilized antimicrobials on treated surfaces. Similar modifications have proven successful in determining viral efficacy of the active found in Armatrex against similarly structured lipophilic enveloped RNA and DNA viruses.
- We should discuss expanding the list of viruses to include swine viruses such as *PEDv* and *Betaarterivirus suid 1 formerly known as PRRSv*. We can look at Poultry viruses and Companion animal viruses as well.
- Clearstream is working on a new modification of an existing American Society for Testing and Materials (ASTM) standard test that will combine the current ASTM protocols and protocols derived from viral testing

performed at the University of Michigan (*Wang et al.*) against the active in Armatrex. Clearstream anticipates this new version could become a standardized method for testing immobilized antimicrobials against viruses.

# Ohio State University (Food Science Department) Microbial Challenge/Delayed Inoculation Challenge/Durability Challenge

- Multiple Stainless steel treated, and control sample coupons were provided to the Food Science Department at OSU to conduct a series of studies providing evidence of Armatrex and the Clearstream suite of branded products efficacy against two common food borne bacteria at various time periods and surface challenges.
  - Upon receipt of the coupons the first microbial challenge was conducted.
  - Additional coupons were put in storage for a period of three (3) months to delay the treated coupons from being subjected to a microbial challenge.
  - A third group of coupons that were stored to delay the microbial challenge were removed from storage and additionally subjected to vigorous surface challenges mimicking cleaning and scrubbing activities. These coupons were then subjected to a microbial challenge post surface cleaning activity.
- In all three microbial challenges the treated stainless steel coupons exhibited significant efficacy against the two food borne bacterial pathogens, one a gram-negative and the other a gram-positive bacteria as further outlined in the following statements.
- The Clearstream suite of aligned branded products were tested at Ohio State University (Food Science Department) and proven effective against *Listeria innocua* at multiple contact times utilizing a version of the standardized JIS Z 2801 test or the ISO 22196 over an extended period of several months and surface challenge conditions.
- The Clearstream suite of aligned branded products were tested at Ohio State University (Food Science Department) and proven effective against *Escherichiacoli (E. coli)* at multiple contact times utilizing a version of the standardized JIS Z 2801 test or the ISO 22196 over an extended period of several months and surface challenge conditions.

# **Field Studies**

#### South Georgia Broiler House Field Trial

- In June of 2018 Clearstream was asked to look into what it could do to mitigate concerns of area poultry growers in the immediate aftermath of the cessation of daily antibiotic dosing. Clearstream arrived after the results were collected from the first post antibiotic flock cycled through. As background, these chickens were housed for approximately 60-62 days and were expected to weigh in the 8-9 pound range at delivery. In most cases the typical antibiotic cycle mortality rates tripled on average for the non-antibiotic flock versus the previous flocks that were being dosed on a daily basis. Overall weights also showed reductions in the post antibiotic flocks.
- In total there were 4 independent farms in the region with a total of 16 broiler houses that were selected for treatment. Two of the participating farms were selected for repeated treatments for their next placement cycle for a total of 24 cycles.
- In all cases, historical data was collected from daily sheets and integrator summaries for mortality, weights, and overall viability. This data was chosen from the two most recent cycles prior to cessation of daily antibiotic dosing and the initial non-antibiotic cycle that immediately followed the prior daily dosing cycles. This gave Clearstream the proper benchmark to determine the efficacy of Clearstream's antimicrobial and its performance value relative to the existing non-antibiotic conditions and the prior daily antibiotic dosing conditions.
- The 4 participating farms presented a wide set of physical conditions within the structures themselves as well as how the growers approached the preparation of the houses in advance of their next chick placement. Age of structures, overall conditions of the structures, and attention to specific issues relating to litter prep, ammonia control, and Darkling beetle pest controls were all noted.
- None of the 16 houses in this trial had a full clean out or litter replacement prior to Clearstream's treatments. The only consistent

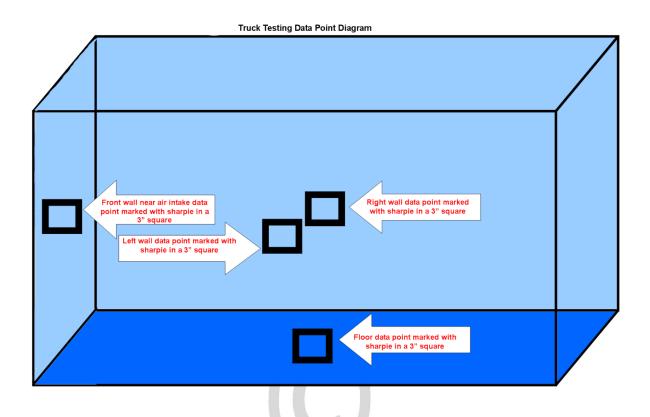
litter preparation that was observed on all 4 participating farms was the removal of the cake layer and smoothing out of the litter using a drag harrow. No interior surfaces or appurtenances were blown, power washed or cleaned in any manner prior to Clearstream's treatments. It was revealed in all instances that the litter in the participating broiler houses had been in place on average for 24 months.

- Due to the short notice and the urgency that was displayed, Clearstream's initial treatments on the first participating farm were performed by two applicators each utilizing a handheld backpack electrostatic sprayer.
- In subsequent return visits the spray equipment that was utilized increased in sophistication culminating with a PTO driven multiheaded sprayer cutting down time immensely and increasing the uniformity of coverage.
- In every treatment event Clearstream treated substantial expanses of existing substrates that were heavily laden with bioburden. Walls, ceilings, and litter were treated one (1) time approximately 4 to 7 days prior to bird placements.
- At the culmination of the trial all the data was collected, tabulated, and analyzed. Feed Conversions improved, overall viability improved, and mortality rates were reduced. The percentages of mortality reduction were compared most directly to the first non-antibiotic flocks on the 4 participating farms that our treatments followed. On the low side we observed mortality reductions of 34% and on the high side of 52.5% The average mortality reduction was 43.6%. Weekly percentages in various houses displayed mortality reductions over the previous non-antibiotic flock as high as 78% and 84%. These weeks were closer to the beginning and middle of the cycles.
- In summary, Clearstream theorized that the greatest improvement was experienced during the first 7 days and in the immediate weeks that followed that initial period. Chicks weigh less, and are walking on top of the semi-compacted, but treated litter. This buffers the chicks initially and gives them a head start by allowing their immune systems to get up to speed without the overwhelming contamination loads present in the buildings, specifically in the highly contaminated litter.
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• As the treated litter becomes increasingly burdened with excrement from the maturing birds, Clearstream's antimicrobial will lose its ability to perform as the molecules are covered in layers of gross filth, but theoretically it doesn't play as significant a role at that time as the young birds have hopefully benefited from a healthier and safer first seven (7) day period and the immediate period that follows..

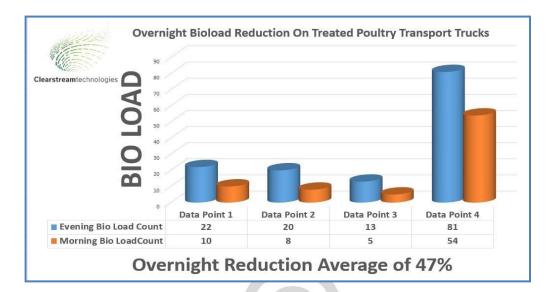
# South Alabama Chick Delivery Truck Field Study

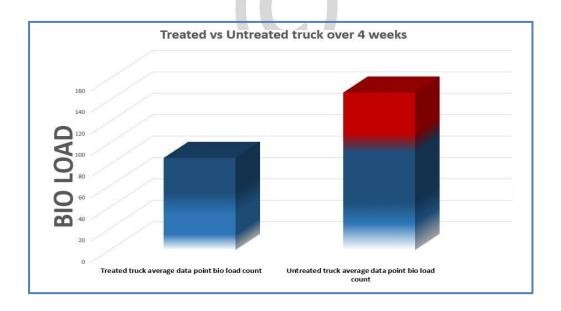
- From July 1, 2015 to July 22, 2015 Clearstream Technologies, in conjunction with a major global poultry breeder performed a field trial to track the performance of Clearstream's residual antimicrobial on poultry transport trucks at the breeder's Alabama facility.
- The purpose of this trial was to compare the bioloads of 2 treated trucks with Clearstream's residual antimicrobial and 3 similar untreated trucks; and to determine the efficacy of the antimicrobial in the reduction of biolevels between surface cleanings.
- The following pages report the data retrieved during the period outlined above. The data was collected using Hygiena Ultra Snap Swabs and a standard Hygiena Adenosine Triphosphate (ATP) Bioluminescence Luminometer. All data was collected and recorded onto an excel spread sheet for the purposes of comparison and verified by a breeder employee. The following pages contain all of the raw data collected and a brief written summary.
- The data sites of this study were comprised of 4 data points per truck) as shown in the illustration below. Each data site was, catalogued and recorded onto an excel spread sheet after the truck was used for a delivery of live chicks.



- The treated trucks of the study site were originally cleaned and disinfected with an EPA registered quaternary biocide Detergent/Disinfectant and then immediately treated with Clearstream's residual antimicrobial.
- The untreated trucks were cleaned using the breeder's standard protocols.
- The tests were taken at 3 specific times of day:
  - o upon the return of an empty truck after delivery,
  - $\circ$  the next morning before disinfection and
  - $\circ$  post disinfection.
- Due to complications in breeder's scheduling some of the data collection was inconsistently recorded.
- These inconsistencies in data collection were eliminated in the overnight summary as not to skew the actual results.
- The subsequent graphs represent the average overnight bio load reductions where the data was collected consistently within the guidelines of the collection protocol with the correct comparison of bio load build up between treated and untreated trucks.

• The antimicrobial consistently reduced the bio load on all data points, slowed colonization and reduced bio load buildup.





Conclusion

- During this trial, the breeder proved to be maintaining a respectable disinfection protocol.
- Even with good disinfection and cleaning, it is difficult to maintain consistently low bacterial and contamination levels due to the propensity

for organic grime layers to build up rapidly in poultry transports.

- This reality contributes to potentially high levels of bacterial, fungal, and viral threats.
- Even in this abbreviated study Clearstream's residual antimicrobial demonstrated that regular treatments would reduce the threat to newborn chicks first 7 day mortality rates.

# Southern Tennessee Chicken Breeder House Field Study

- A Clearstream version of the Armatrex antimicrobial was deployed in a poultry breeder house in November of 2014 and was compared to its adjoined untreated control house. The entire house including nesting boxes and belts were treated one (1) time, post clean out, and just days prior to the placement of the pullets. At the age of 28 weeks the eggs were collected from the first belt run and were tagged from the treated and control houses. By design, the collected eggs were "overnight eggs" that cooled and were exposed to the conditions present in the houses versus the collection of freshly laid warm eggs.
- The tagged eggs were delivered to the hatchery and upon their hatch the chicks were marked again to differentiate the birds from the treated house and the birds from the control house.
- In summary, at the end of the 1<sup>st</sup> seven days post hatch, the chicks that came from the eggs collected from the treated house had an improved mortality rate of .63% and an improved weight of 2.6 grams over the control house collected eggs.
  - While these results only demonstrated marginal gains, this study along with additional studies that came after helped provide the basis for Clearstream's contention that whole house treatments would ultimately lead to better hatch rates, lower mortality rates, greater feed to weight ratios, and overall stronger flock viability.
  - With greater treatment refinements, new studies in broiler houses, and direct egg treatments which had not yet been conducted, but were being discussed with leading industry veterinarians, a clear direction was defined for the use of Clearstream's residual antimicrobial technology.

# South Carolina Hatchery (Hatcher) Durability Field Study

- A Clearstream version of Armatrex was used to treat 28 poultry hatchers in a South Carolina hatchery. Over a period of nine (9) months Clearstream revisited the hatchery and tested for the presence of the active found in Armatrex on the treated surfaces.
- After only one (1) treatment and approximately 60+vigorous cleaning and disinfecting procedures that included power washing, the use of a powerful Quat/Glutaraldehyde disinfectant and scrubbing with Teflon style pads, the active in Armatrex was still found on surfaces in every hatcher tested at the end of the 9<sup>th</sup> month.

# West Texas Dairy Calf Hutch Field Trial

- A joint study was conducted on the premises of a working dairy farm operation in West Texas to determine the efficacy of Clearstream's antimicrobial treatment protocol to dairy calf hutches.
- The test results of the bio load accumulation on the hutches cleaned, disinfected and treated with an EPA registered biocide and Clearstream's biostatic antimicrobial products were compared and contrasted to the test results of the bio load accumulation on the hutches cleaned, disinfected and treated with the existing dairy sanitization protocols at agreed upon intervals during the study.
- The study was jointly undertaken by Clearstream Technologies, LLC ("Clearstream"), Purina Animal Nutrition, LLC ("Purina"), and a wellestablished dairy management team beginning on September 16, 2014. The final report is a collaborative effort of all of the foregoing stakeholders.
- This study commenced when a row of one hundred (100) contiguous hutches became available on the dairy.
- For the purposes of identification during this study, the back of the Clearstream hutches (hereinafter referred to as the "Test Group") were marked with green masking tape and numbered 1 50 on the face of the tape.

- Likewise, the back of the hutches to be cleaned and treated using the existing dairy protocol (hereinafter referred to as the "Control Group") were marked with red masking tape and numbered 1 50 on the face of the tape.
- Purina Dairy Calf and Heifer Specialists Dr. Christie Underwood, Ph.D., and Bethany Fisher, M.S. - performed testing to determine bio load levels present on the interior surfaces of the subject calf hutches using a Hygiena SystemSure Plus ATP detection meter and retrieved test swabs on each of the fifty (50) hutches in the Test Group prior to any cleaning activities commencing as a baseline and control measure.
- These readings provided an indication of the average amount of bio burden that accumulated over the duration of the prior set of dairy calves' occupation of the same hutches chosen for this study.
- ATP test swabs were not taken on any of the fifty (50) hutches in the Control Group prior to being cleaned and disinfected using the existing dairy sanitization protocol because these hutches had been cleaned by dairy personnel prior to the arrival of the Clearstream and Purina personnel.
- Shortly after each of the one hundred (100) hutches had been cleaned, ATP swabs were taken by Purina personnel on all one hundred (100) hutches involved in the study in the same location on each hutch to provide a second baseline and control measure.
- These readings provided an indication of the bio burden that existed on the inside of each hutch after cleaning and disinfection and prior to the introduction of the calves into the hutches the subject this study.

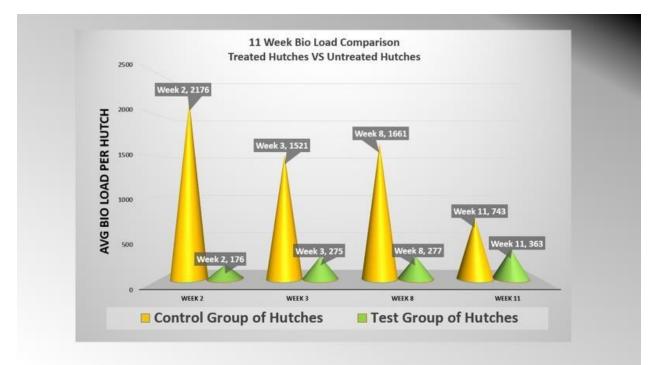
#### In Brief:

# Dairy Calf Hutch Study Conducted with Purina Animal Nutrition:

- 82 day study was conducted on a large dairy farm in West Texas beginning September 14, 2014.
- Study included 100 calf hutches. 50 hutches were control hutches and 50 hutches were test hutches.
- Test hutches were cleaned, disinfected, and treated only once with our residual antimicrobial product at the onset of the trial period.
- · Control hutches were cleaned and prepared with existing protocols.
- A group of the test hutches were selected to receive a weekly disinfecting soil amendment.
- Bio accumulation in calf hutches are typically significant. Open air, humidity, dust, urine, feces, and mucous are the main contributors.

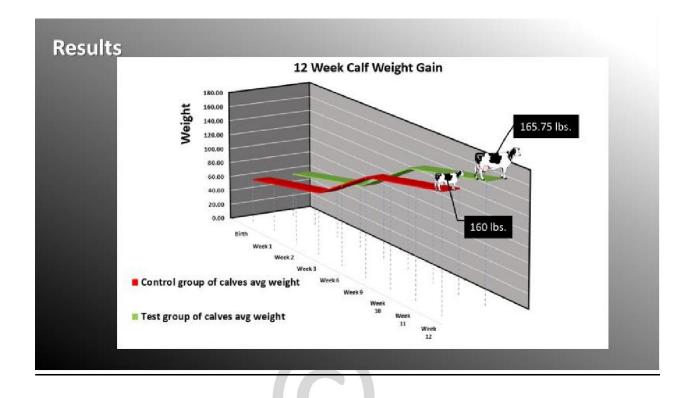
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# Dairy Calf Hutch Study Conducted with Purina Animal Nutrition:

- Calves in the test hutches were exposed to 92% less bio burden in week
   This is a critical time in the young calf's life and the test hutches displayed a significantly improved living condition over the control hutches.
- Calves in the test hutches were exposed to between 82 to 84% less bio burden between weeks 3 and 8 over the control hutches.
- After 82 days the calves in the test hutches were exposed to 51% less bio burden than the control hutches.
- Calves in the test hutches gained an average of between 3.28 pounds (.04 pounds per day x 82 days) and 5.74 pounds (.07 pounds per day x 82 days with weekly soil amendment) greater than the average weight gained by the control group.



# **Poultry Lab Studies**

#### University Of Georgia Egg and Chick Study Laboratory Trial

- The study was conducted by Dr, Jeanna Wilson and Laura Oxford at University of Georgia, Poultry Science Department, February 18-March 19, 2019 and sponsored by Dr. Ivan Alvarado of MAH.
- The study was titled "Evaluation of the Effect of Electrostatic Aspersion of Armatrex, a Novel Silane Quaternary Ammonium Compound (QAC) on Fertile Eggs Hatchability and Chick Quality"
- Naturally occurring bacteria in a commercial hatchery can be detrimental to hatchery performance as well as chick health even though sanitation and disinfection occur routinely.
- Additionally, as the poultry industry moves to antibiotic free production, there is concern that hatcheries will become a source of bacteria to chicks. Because of this, novel sanitation processes are being tested. These processes include eggs being treated directly with antimicrobial chemicals.

#### **Experimental Design:**

- The electrostatic application of the Armatrex silane quaternary ammonium was at 0.1, 0.25, 0.5, and 1% of the active ingredient by volume (approximately 540 eggs per treatment or 6 full trays of eggs per treatment). These levels were compared to the control non-treated eggs.
- The broiler eggs used in this study were obtained from a commercial hatchery and were from a 31 week old flock from one day's egg collection with the same storage conditions.
- The eggs were candled at 12 days of incubation and the clear and early dead eggs removed.
- Water vapor conductance, moisture loss, residue analysis, chick quality and porosity were evaluated in this study.

# The Study Results are Displayed in the Following Tables:

**Table 1.** Average water vapor conductance from eggs randomly selected from each treatment group.

Average Water VapoTRTConductance(mg H2O/d/Torr/100 g)						
Control	17.09					
0.1%	14.67					
0.25%	17.60					
0.5%	21.03					
1.0%	17.96					



**Table 2.** Percentage of moisture lost from a random selection of eggs from each treatment group.

TRT	% Moisture Loss
Control	10.96
0.1%	10.75
0.25%	10.82
0.5%	10.64
1.0%	10.72

TRT	% Hatch	% Hatch of Fertile	% Early Dead	% Middle Dead	% Late Dead	% Live Pip	% Dead Pip	% Live Cull	% Dead Cull
Control	85.5	86.81	6.88	0.00	4.09	0.56	0.37	0.19	0.00
0.1%	87.93	88.43	4.63	0.19	3.16	0.56	1.31	0.56	0.56
0.25%	90.56	91.41	2.96	0.37	3.33	0.93	0.37	0.19	0.37
0.5%	87.59	88.74	6.48	0.19	2.59	0.19	0.74	0.00	0.74
1.0%	85.19	86.27	6.67	0.00	4.26	0.19	0.37	1.30	0.37

**Table 3.** Incubation data collected from candling, hatch, and residue analysis.*One-way ANOVA* 

Early dead p-value(between 0.25% and control) = 0.0726Live cull p-value (between 0.5% and 1.0%) = 0.0532

**Table 4.** Average yolk weights and yolk-free body weights after hatch.

TRT	Yolk Weight (g)	YFBW (g)
Control	4.07	33.36
0.1%	4.36	33.98
0.25%	3.9	33.67
0.5%	3.95	33.95
1.0%	3.99	34.16

**Table 5.** Average chick weights at hatch per treatment.

TRT	Average Chick Weights (g)
Control	38.07
0.1%	38.58
0.25%	36.38
0.5%	38.54
1.0%	38.98

 $\bigcirc$ 

Pen Number	TRT	Pen Weight (kg) - Day 1 March 12	Pen Weight (kg) - Day 7 March 19	Adjusted Gain (kg)
1	Control	0.765	1.535	0.77
2	0.10%	0.75	1.705	0.955
3	0.25%	<b>25%</b> 0.74 1.4		0.712
4	0.50%	0.765	1.675	0.91
5	1.00%	0.775	1.675	0.9
6	<b>Control</b> 0.755 1.56		1.56	0.805
7	0.10%	0.74	1.57	0.867
8	0.25%	0.745	1.585	0.84
9	0.50%	0.76	1.675	0.915
10	1.00%	0.76	1.71	0.95

Table 6. Pen weights at placement and termination.

\*1 mortality in pens 3 and 7 = 1%

**Table 7.** Mycology results from lung samples.

	Percent Positive Chicks							
TRT	Aspergillus spp.	Penicillium spp.						
Control	50	10						
0.10%	20	0						
0.25%	10	0						
0.50%	10	0						
1.00%	20	0						

\*Necropsy findings from each treatment group were grossly normal. Samples for bacteriological evaluation from the yolk sacs and organ pools of each treatment resulted in no growth.

#### In Conclusion:

- In nearly every category the Armatrex treated eggs were within acceptable margins for water vapor conductance and moisture loss or they outperformed the control eggs with higher percentages of hatch and fertile hatch, hatch weights, and pen weights.
- Most importantly the Armatrex treated eggs dramatically reduced the presence of *Aspergillus* by 60 to 80% compared to the control eggs and eliminated *Penicillium* completely over the control hatched chicks across all of the treatment concentrations.

- Of great importance, in almost every category the 1% active concentration in Armatrex performed very well and proved to be very safe.
- Lastly, as shown above in the **Treated Substrate Observations**, the Armatrex antimicrobial across all treatment levels demonstrated no pore obstruction on any of the shell fragments.
- In both the dyed shell fragments imaged by the Light Electron Microscope and the subset of fragments imaged by the Scanning Electron Microscope it is clear that Armatrex poses no inherent risk to the embryos, but instead by virtue of the above test results appears to be quite beneficial to the overall health of the embryo and hatchling.

# Healthcare and Commercial Food processing

# Summaries of Multiple Hospital ATP Field Testing and Commercial Meat Processing Plant (USDA Listeria Challenge & Mitigation) Treatment

## Fredrick Memorial Hospital/Calvert Medical Center (Maryland)

Clearstream Technologies in conjunction with Compass Crothall Environmental Services (EVS) staffs at Calvert Health Medical Center in Prince Fredrick, MD and Fredrick Memorial Hospital in Fredrick, MD conducted a controlled sixty (60) day study. The study was designed to gauge the effectiveness of Clearstream's Penetrexx immobilized (static) antimicrobial's introduction into the existing cleaning and disinfection protocols at both hospitals simultaneously.

- The study commenced at both hospitals on June 21, 2017 and continued for 60 days.
- The purpose of the testing was to prove continued efficacy of Clearstream's Penetrexx antimicrobial and the reduction of bioburden on treated and control surfaces versus normal EVS cleaning and disinfection protocols utilizing Adenosine Triphosphate (ATP) swabbing and metering.
- ATP metering scores are widely accepted and utilized in a growing number of hospitals, healthcare centers, food manufacturing and processing facilities and restaurants as a means of monitoring how EVS personnel are achieving the established mandates set by the epidemiology and infectious disease departments of individual hospitals and healthcare systems and food manufacturers and providers.

- The recorded results clearly indicate that the Penetrexx static antimicrobial remained efficacious throughout the 60-day trial period with only one application at the onset of the trial.
- Penetrexx performed as expected and the collected ATP scores reflect this fact by way of a significant differential in ATP count reductions on treated surfaces versus untreated control surfaces.

# The Treatment & Test Trial Sequence:

- In each hospital, five (5) high volume touch surfaces were chosen in an occupied patient room identified and chosen by the EVS staff and treated (Treated Surfaces). Each EVS staff further identified occupied patient rooms within each hospital facility and five (5) identical surfaces were designated for testing, but were untreated as (Control Surfaces)
- 10 surfaces in total were identified as 5 treated and 5 control.
- The untreated surfaces were designated the control ("C") and the treated surfaces were designated the test ("T"). The surfaces included a patient room entrance door handle, toilet handle, TV handle, soap dispenser handle and TV remote.
- ATP test swabs were taken on all ten (10) identified surfaces as a pretrial baseline prior to any cleaning activities performed by Clearstream.
- The test room surfaces were disinfected with an EPA registered QAC disinfectant and treated with Clearstream's Penetrexx static antimicrobial.
- The treated surfaces were marked and designated as the "Test Group" and documented with photographs.
- The remaining 5 surfaces were not treated by Clearstream. They were marked and designated as the "Control Group" and documented with photographs.
- Immediately after Penetrexx had dried and partially cured, ATP test swabs were taken on the 5 treated surfaces to establish a baseline and act as a control.
- ATP swab testing of the "Control Group" and "Test Group" were performed two times per week at intervals as follows:
  - Testing commenced the morning after treatment and was performed two (2) times per week on "test and control" surfaces in the early morning prior to regularly scheduled

facility cleaning protocols and continued for a period of 60 days.

- A second test was performed two (2) times per week at the end of the (normal hospital business) day after the "test and control" surfaces were exposed to normal daily use and continued for a period of 60 days.
- All swab test results were recorded as taken by EVS staff on master data sheets.
- All surface touch locations were swabbed in the exact same locations at each time interval to produce a consistent sampling and analysis.
- All the collected data sets were compiled on Excel Spread Sheets on a weekly basis and compiled at the end of the 60 day trial to compare the statistical performance of occupied patient room surfaces that had the one time treatment of the Penetrexx static antimicrobial introduced into the protocol versus the untreated occupied patient room control surfaces that were monitored during the trial period.

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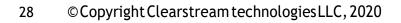
<u>The Following Graphs Help Illustrate The Significant Differential Between</u> <u>The Treated And Control Surfaces</u>.

**(C)** 

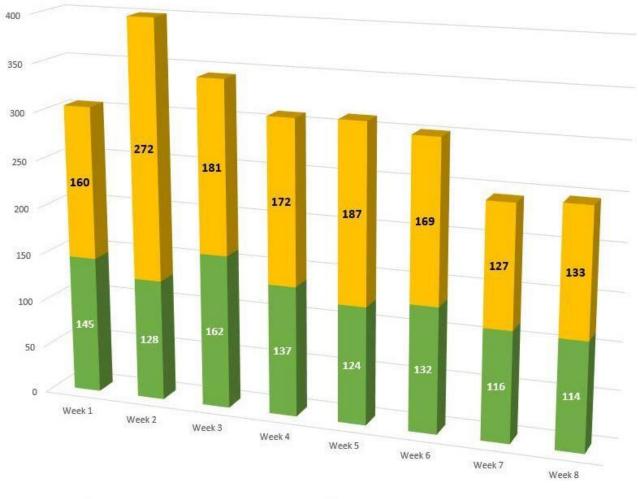
**(C)** 

**Calvert Health Medical Center** 

(C)



#### **Fredrick Memorial Hospital**



Treated Room (test) ATP SCORE Untreated Room (control) ATP SCORE



Ireated Room (test) ATP SCOR

## St. Elizabeth Hospital, Cincinnati, OH

The following are results compiled from two separate field trials (Phase 1, Phase 2) conducted at St. Elizabeth Hospital in Cincinnati, OH in December of 2019 and January of 2020.

## Phase 1

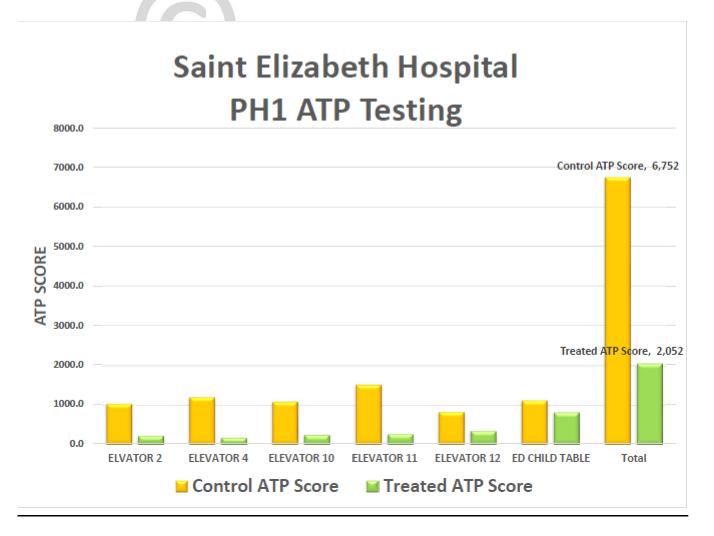
- Phase 1 testing was done on call buttons in a series of elevator banks and a child's table which are high volume touch surfaces.
- The untreated test surfaces were maintained with existing EVS cleaning and disinfection protocols and swabbed every day for 5 concurrent days.
- The ATP readings were tabulated prior to treatment.
- The surfaces were cleaned again and then treated with our Penetrexx immobilized (static) antimicrobial technology and were swabbed for an identical concurrent 5 day span post treatment.
- The ATP readings were tabulated and compared to the previous 5 day control period. All surfaces were maintained with existing protocols by their staff pre (control) and post treatment. These results showed a consistent 4 to 1 improvement ratio in ATP scores after one treatment over the untreated control surfaces with a very high volume of activity.

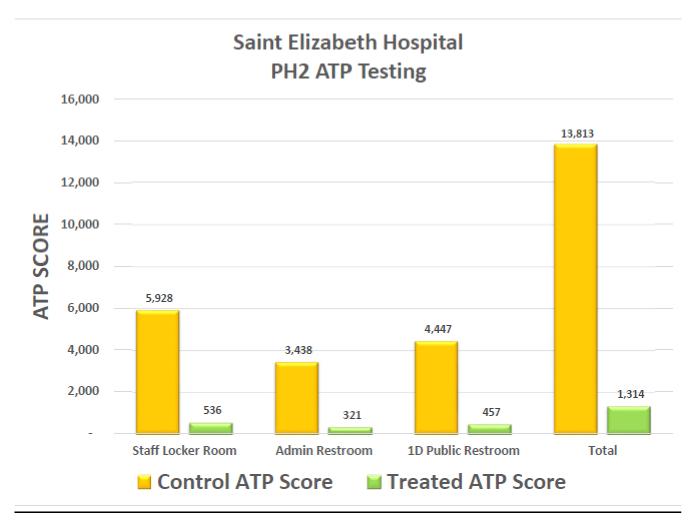
## Phase 2

- Phase 2 testing was conducted similarly with specific lavatory surfaces that were chosen by EVS personnel for consistent activity including seats and door handles.
- The surfaces were maintained by the existing EVS protocols during the control period which was run over 6 consecutive days with daily ATP swabbing's.
- The ATP readings were tabulated.
- The surfaces were cleaned again and treated with the Penetrexx static antimicrobial to start the next consecutive 6 day period.
- Daily ATP swabbing's were conducted, and the readings were tabulated.
- These results showed a nearly 15 to 1 improvement ratio of ATP scores after one treatment over the control surfaces with consistent activity levels.

All decisions on what surfaces were chosen for testing, physical treatments of the test surfaces, ATP swabbing, data retrieval and tabulating were performed by the Epidemiology department and Environmental Services personnel (Xanitos EVS) at St. Elizabeth's Hospital.

The Following Graphs And Tables Will Help Illustrate The Significant Differential In ATP Scores Between The Treated And Control Surfaces







Phase 1	D/	(Y 1	D/	AY 2	DA	¥ 3	D/	AY 4	D/	N 5			To	tals		
Surface	Control ATP Score	Treated ATP Score			Control ATP Score	Treated ATP Score	Percent reduction	RATIO								
levator 2	19	167	306	51	1,424	287	1,724	301	1,674	293			5,147	1,099	468%	4.68 to 1
levator 4	256	126	504	122	1,947	134	1,550	223	1,725	251			5,982	856	699%	6.99 to 1
levator 10	135	30	466	313	1,590	294	1,610	281	1,605	293			5,406	1,211	446%	4.46 to 1
levator 11	157	98	844	295	4,024	304	1,205	316	1,324	308			7,554	1,321	572%	5.72 to 1
levator 12	278	458	422	306	1,130	322	1,145	315	1,135	302			4,110	1,703	241%	2.41 to 1
D Child table	424	814	733	815	1,418	820	1,507	810	1,478	813			5,560	4,072	137%	1.37 to 1
fotal	1,269	1.693	3.275	1.902	11,533	2.161	8,741	2.246	8,941	2.260			WG Percer	t Reduction	427%	4.27 to
	-,									_						
Phase 2	DA	W 1	DV	AY 2	DA	Х 3	D/	AY 4	DA	N 5	DA	Y 6	То	tals		
Surface	Control ATP Score	Treated ATP Score	Percent reduction	RATIO												
taff Locker room																
tall 1 toilet seat	47	8	34	29	75	2	136	0	96	32	843	16	1231	71	1734%	17 to 1
tall 2 toilet seat	153	31	103	17	763	0	1569	7	260	19	426	10	3274	74	4424%	44 to 2
tall 3 toilet seat	5	22	13	40	88	20	60	3	120	26	4	18	290	111	261%	3 to 3
tall 4 toilet seat	124	39	48	7	175	9	48	0	88	15	186	22	669	70	956%	10 to 4
oor handle	76	37	110	39	62	15	30	12	120	16	66	25	464	119	390%	4 to 5
idmin restroom														••		
tall 1 toilet seat	268	24	258	0	655	32	39	7	225	31	28	20	1473	94	1567%	16 to 5
tall 2 toilet seat	40	12	193	5	919	19	17	4	182	12	250	14	1601	52	3079%	31 to 5
oor handle	17	7	208	6	12	28	34	23	48	47	50	30	364	111	328%	3 to 5
D public restroom								-			-					
tall 1 toilet seat	122	30	492	6	118	17	260	13	111	37	256	19	1359	103	1319%	13 to 5
tall 2 toilet seat	249	2	51	1	19	17	129	9	151	58	483	30	1082	87	1244%	12 to 5
itall 3 toilet seat	222	6	212	16	63	1	155	0	123	56	135	25	910	79	1152%	12 to 5
Door handle	169	0	283		63	24	51	20	43	41	487	22	1096	92	1191%	12 to 5
fotal	1,492	218	2,000	173	3,012	184	2,528	98	1,567	390	3,214	251	VVG Percer	nt Reduction	1470%	14.70 to:

#### St Elizabeth Hospital ATP Data PH 1 and 2

#### LA Vinas M.D., Boca Raton, FL

Clearstream Technologies, LLC., in conjunction with the medical practice of Dr. Luis A. Vinas M.D., performed a study of 20 high volume touch surfaces at Vinas Plastic Surgery and Medical Spa in Boca Raton, FL.

- The purpose of this study was to compare the bio loads of 20 surfaces treated with Sabretech QS and 20 similar untreated surface; and to determine the efficacy of Sabretech QS in the reduction of bio levels between surface cleanings.
- The following summarizes the reported data retrieved during the study period.
- The data was collected using Hygiena Ultra Snap Swabs and a standard Hygiena ATP Bioluminescence Luminometer. All data was collected and recorded onto an excel spread sheet for the purposes of comparison.

### Summary

- The data sites of this study were comprised of a range of surfaces including medical equipment, door and cabinet handles, desks, hampers, and sinks which were used by the doctor and his staff on a daily basis.
- Each site was clearly marked as to what would be treated (Treated/Green) and what would be untreated (Control/Red) as shown in the illustration below. Each data site was photographed, catalogued and recorded onto an excel spread sheet.
- The treated side of the study site was originally cleaned and disinfected with an EPA registered QAC disinfectant and then immediately treated with Sabretech QS biostatic antimicrobial.
- The untreated side of the site was cleaned with the EPA registered QAC disinfectant to create a comparable starting point as the treated side.
- The tests were taken at two specific times of day; at the close of business to effectively show the bio-loading of surfaces during use of the offices and the following morning prior to opening of the offices to demonstrate Sabretech QS's efficacy with no outside interaction or cleaning of those surfaces prior to the morning testing.
- The most significant data was taken on the last two days of the study. Data was collected after the facility closed on and again before the facility opened.
- While comparing the average reduction on all data points between these two times, the data points treated with Sabretech QS contained 240% less bio-load than the untreated data points, or were 763% more effective at reducing bio-loads than untreated surfaces.
- This is a significant difference when considering the test surfaces had been heavily used for a 6 week period of time post Sabretech QS application. Even rigorous daily use of treated surfaces did not remove the Sabretech QS molecule or prohibit it from performing at a high level.
- Considering the data collection over the entire 6 week period. On
- 34 © Copyright Clearstream technologies LLC, 2020

average, the surfaces treated with Sabretech QS contained **161%** less bio-load and showed overnight reductions in bio-load upwards of **613%** when compared to untreated control surfaces.

• During the 6 weeks of testing the cleaning of equipment, desk tops, sinks and handles was extremely random as was the introduction of bacterial and viral levels due to the everyday use of the office. On average the Sabretech QS treated test sites maintained a significantly lower biological count than the untreated sites.

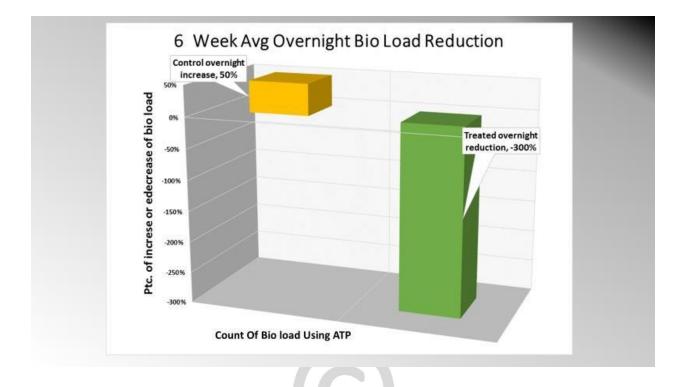
#### The Following Slides Help Simplify The Study

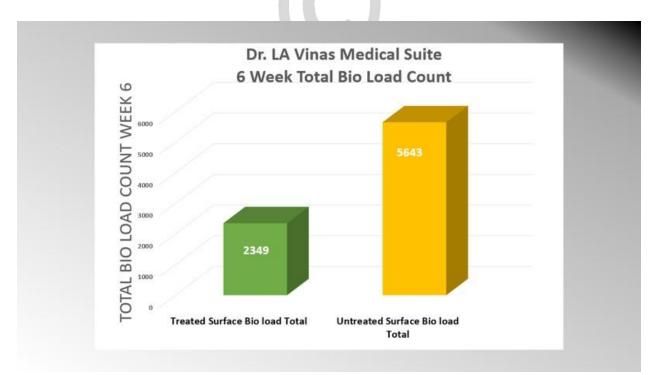
# Dr. LA Vinas Six Week Study of SiQuat Efficacy Against Bio Loads on Medical Suite Surfaces

- Study was conducted over six consecutive weeks
- 40 data points were selected
   20 treated (test) and 20 untreated (control)
- ATP swab samples were obtained between 6pm and 7pm once per week on treated and untreated control points and then again the following morning between 6am and 7am
- · These samples were then compared to one another
- The following graph shows the average percentage of growth and average percentage of reduction of bio load for the control and test data points respectively over the entire six week period



Example of Data Points





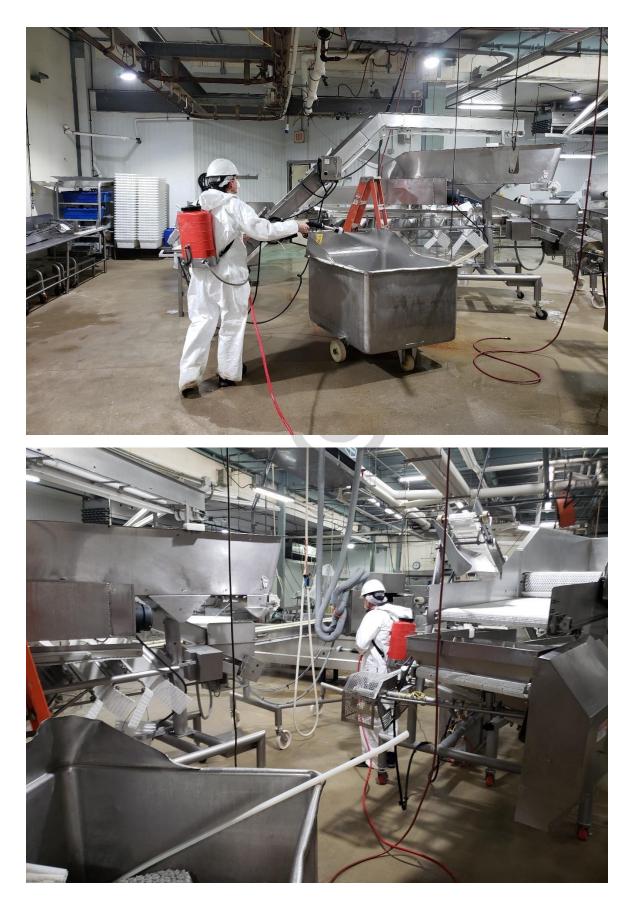
# Established Meat and Meat Product Manufacturer in Southwest Ohio

In Mid-February 2020 Clearstream, through one of its distributors was asked to get involved with a USDA inspection problem that one of their food processing customers had recently experienced in their processing plant in Southwest Ohio.

- Clearstream was informed that this processor had failed a recent USDA inspection for the presence of *Listeria monocytogenes* in multiple locations within the plant and subsequent to those findings the plant was shut down.
- Clearstream agreed to assist the plant management and consulted with senior officials on a preferred disinfection protocol.
- The protocol called for a dual pronged approach utilizing EPA registered applications of a peracetic acid, rinse, and a follow up application of a dual chain quaternary ammonium compound disinfectant.
- These steps were followed by plant personnel prior to Clearstream's arrival. Upon inspection of the plant and its equipment Clearstream agreed that it was ready for the Biotrexx 247 antimicrobial treatment.
- Clearstream made application to the entire facility including wall, ceilings, floors, internal and external processing equipment surfaces, lavatories, offices, etc.
- The facility was approximately 20,000 square feet under roof. Clearstream used approximately 5 gallons of product for the entire treatment.
- The next day the USDA returned for their follow up inspection. USDA inspectors plated 30 separate sample areas including conveyor belts, hoses, internal and external equipment parts, hoses, floors, walls, door handles, and employee shoes.
- Four (4) days post inspection the USDA presented their findings to the plant's senior management. No *Listeria* was found in any of the 30 plated samples. Management was cleared for reopening 2 days after that.
- The plant management now utilizes Clearstream's Biotrexx 247 antimicrobial as a post disinfection application.
- Through multiple USDA inspections since their reopening no *Listeria* has been found and the plant continues to run uninhibited.

# **Treatment Photos at Processing Plant**





# **Toxicity Testing**

### In-Vitro Laboratory Oral Toxicity Testing

- In September and October 2017 Clearstream commissioned an oral toxicity study by the Institute for In-Vitro Sciences (IIVS).
- The study "MTT TIME COURSE ASSAY USING THE EPIORAL<sup>™</sup> MODEL: SCREENING PROTOCOL"
- The EpiOral<sup>™</sup> Model was used to assess the potential oral irritation of the test articles. The MTT (3[4,5-dimethylthiazol-2-yl]-2,5-diphenyltetrazolium bromide) conversion assay, which measures the NAD(P)H-dependent microsomal enzyme reduction of MTT (and to a lesser extent, the succinate dehydrogenase reduction of MTT) to ablue formazan precipitate, was used to assess cellular metabolism after exposure to a test article for various exposure times.
- The duration of exposure resulting in a 50% decrease in MTT conversion in test article-treated EpiOral<sup>™</sup> tissues, relative to control-treated tissues, was determined (ET50 value).
- The purpose of this study was to evaluate the potential toxicity of the test articles to be used as both controls and treated articles (SABRETECH 20FC), supplied by Clearstream Technologies, as measured by the conversion of MTT by EpiOral<sup>™</sup>tissues after exposure to a test article for various exposure times.
- The laboratory phase of the study was conducted from 29 August 2017 to 31 August 2017 at the Institute for In Vitro Sciences, Inc.
- The test articles were tested in a screening assay to determine the duration of exposure to a test article, which resulted in the ET50 endpoint.

## **RESULTS AND DISCUSSION**

## MTT Assay

- The test articles, Lucitone 199 Denture Material treated and Lucitone 199 Denture Material - untreated, were tested in duplicate EpiOral<sup>™</sup> tissues at four exposure times of 4, 8, 16, and 24 hours.
- Table 1 summarizes the ET50 results of the EpiOral<sup>™</sup>assay for the test articles and the positive control. The exposure time response curves are included in Appendix B. The ET50 value for the positive control, 1% Triton®-X-100, fell

within two standard deviations of the historical mean (0.52 to 1.76 hours), thereby meeting the acceptance criteria.

• The test articles, Lucitone 199 Denture Material - treated and Lucitone 199 Denture Material - untreated, were not observed to directly reduce MTT in the absence of viable cells.

Table 1

Assay Date	IIVS Test	Sponsor's	Conc.	ET50	рН
	Article Number	Designation	(w/v)	(hours)	PII
30 August 2017	17AG22/17AG23	Lucitone 199 Denture Material - treated*	Neat	> 24	ND
	17AG24	Lucitone 199 Denture Material - untreated	Neat	> 24	ND
	Positive Control	1.0% Triton®-X-100	NA	1.43	NA

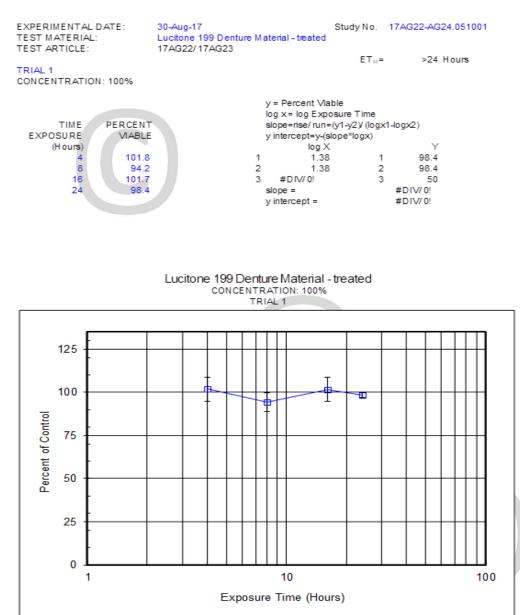
Summary Results of the EpiOral<sup>™</sup> Screening Assay

\* Test article Lucitone 199 Denture Material - treated was prepared according to the protocol (by coating test article Lucitone 199 Denture Material - to be treated with test article SABRETECH 20FC).

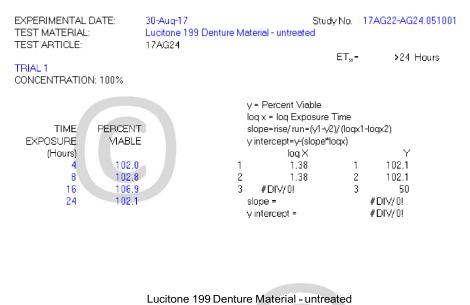
- ND Not Determined; test article was a solid and hence pH value could not be determined
- NA Not Applicable

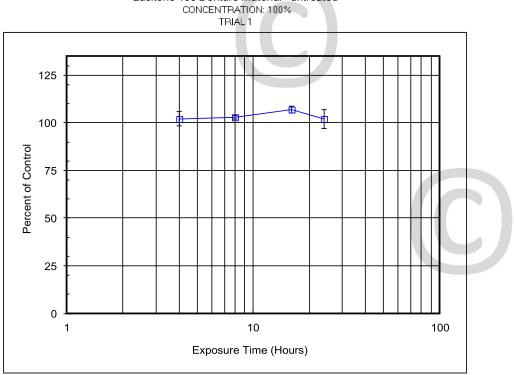


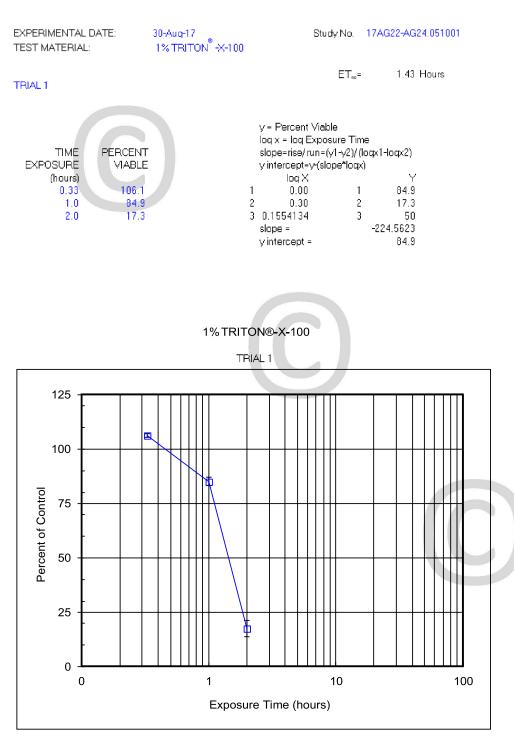
#### EpiOral"BIOASSAY



#### EpiOrall BIOASSAY







EpiOral I BIOASSAY