EVALUATION OF AUTOMATIC CLASS III DESIGNATION (DE NOVO) FOR THE STERIS VERIFY SPORE TEST STRIP FOR S40

REGULATORY INFORMATION

FDA identifies this generic type of device as:

> **SPORE TEST STRIP:** The spore test strip consists of a carrier or strip with a known number of spores, at least 5 $\log_{10}$ per strip, of known resistance to a particular liquid chemical sterilant in a liquid chemical sterilant processing system. A "no growth" result from the spore test strip after the specified pre-determined incubation period indicates that the liquid chemical sterilization process achieved the conditions necessary to kill the specified minimum number of viable spores on the test strip which is 5 $\log_{10}$ spores/strip; it does not confirm the expected full performance of the liquid chemical sterilant processing cycle because full performance is a 6 $\log_{10}$ spore kill in a full liquid chemical sterilization cycle.
>
> **NEW REGULATION NUMBER: 880.6887**
>
> **CLASSIFICATION: II**
>
> **PRODUCT CODE: OVY**

BACKGROUND

DEVICE NAME: Steris Verify Spore Test Strip for S40 for use in the Steris System 1E Liquid Chemical Sterilant Processing System

510(K): K100049

DATE OF 510(K) NSE (NOT SUBSTANTIALLY EQUIVALENT) DECISION: JULY 7, 2011

DATE OF DE NOVO PETITION: AUGUST 01, 2011

PETITIONER CONTACT: **STERIS CORPORATION**
5960 HEISLEY ROAD
MENTOR, OHIO 44060-1834

PETITIONER'S RECOMMENDED CLASSIFICATION: CLASS II

INDICATIONS FOR USE

The Steris Verify Spore Test Strip for S40 is intended to provide users with a means to assess spore kill by the S40 sterilant at its use dilution in the System 1E Liquid Chemical Sterilant Processing System. A "no growth" result from the Steris Verify Spore Test

Strip for S40 after 24 hours of incubation indicates that the liquid chemical sterilization process achieved the conditions necessary to kill at least 1 x 10^5 viable spores (5 logs) on the test strip. The Steris Verify Spore Test Strip for S40 does not confirm the expected full performance of the SYSTEM 1E Liquid Chemical Sterilization Cycle.

LIMITATIONS

Please refer to the labeling for a more complete list of warnings, precautions and contraindications.

DEVICE DESCRIPTION

The Steris Verify Spore Test Strip for S40 consists of a 1 3/8 in x ¼ in filter paper-based strip inoculated with *Geobacillus stearothermophilus* spores and is enclosed in a glassine envelope. The Spore Test Strips are provided with media vials containing a modified tryptic soy broth with phenol red pH indicator, and a transfer clip. The transfer clip holds the Spore Test Strip in a single location during a processing cycle in the System 1E Liquid Chemical Sterilant Processing System (System 1E processor; cleared under K090036) and enables aseptic transfer of the Strip from the processor into the growth media vial. Using the clip, the Spore Test Strip is secured on the available post located in the tray of the System 1E processor. The Spore Test Strip specifications are shown in Table 1.

Table 1. Spore Test Strip Specifications

Value	Spore Test Strip for S40
Population	
At manufacture	≥1.5 x 10^5 cfu*/strip
After wash-off (end of cycle)	≥1.0 x 10^5 cfu/strip
Resistance	
D-value	12-26 sec at 1635 ppm* PAA*
Survival Time	≥38 seconds
Kill Time	≤239 seconds
Reduced Incubation Time	24 hours

*cfu = colony forming units, ppm = parts per million, PAA = peracetic acid

For use, the Spore Test Strip is removed from the glassine envelope and secured in the System 1E processor along with the items to be liquid chemically sterilized. The liquid chemical sterilization cycle is initiated. At the end of the full cycle, the Spore Test Strip is removed and placed into the vial of growth medium for incubation under the specified conditions that will promote spore growth for at least 24 hours. If the media remains red and non-turbid, the user interprets the results as a pass. If the media color turns yellow or is turbid, the user interprets the result as a fail.

The Spore Test Strip is incubated at 55-60ºC for a minimum of 24 hours, but may be incubated for up to 7 days. The shelf life is 12 months when stored at 2-24°C and 30-80% relative humidity (RH) away from sterilizing agents and excessive heat.

Use of the Verify Spore Test Strip for S40 with the System 1E Liquid Chemical Sterilant Processing System

The Verify Spore Test Strip for S40 is intended to be used with the System 1E Liquid Chemical Sterilant Processing System (System 1E), which was cleared under K090036. The System 1E Processor dilutes the S40 Sterilant Concentrate to its use dilution (>1820 mg/L peracetic acid), liquid chemically sterilizes the load during a controlled 6-minute exposure at 46 to 55°C, and rinses the load with extensively treated potable water. The processed load should be used immediately. In the 510(k) submission, K090036, for the System 1E processor and the S40 Sterilant, simulated use testing demonstrated a 6 log kill of spores on devices by the end of the 6-minute liquid chemical sterilization cycle.

Limitations to use of liquid chemical sterilants (LCS) for device sterilization[1] The information available in the literature suggests that sterilization processes based on LCSs, in general, may not convey the same sterility assurance level (SAL) as sterilization achieved using thermal or physical methods. The data indicate that the survival curves for a LCS may not exhibit log-linear kinetics and the shape of the survivor curve may vary depending on the formulation, chemical nature and stability of the LCS. It is not possible to directly measure an SAL at the half cycle point of a LCS cycle as it is with thermal sterilization methods using a Biological Indicator. Therefore, the FDA recommends that LCSs be limited to reprocessing only semi-critical and critical devices that are heat-sensitive and incompatible with other sterilization methods.

Assessing Spore Kill by a Liquid Chemical Sterilant

The Verify Spore Test Strip for S40 is an optional accessory that provides users with a means to test the sporicidal activity of the S40 Sterilant use dilution in the System 1E. A "no growth" result after 24 hours of incubation from the Spore Test Strip holding 1×10^5 cfu of viable spores indicates that the liquid chemical sterilization process achieved the conditions necessary to kill at least 1×10^5 viable spores (5 logs) on the test strip by the end of the cycle. It does not confirm the expected full performance of the liquid chemical sterilization cycle, which kills at least 1×10^6 viable spores.

The System 1E is computer controlled and continually monitors the cycle, including the fill time, exposure time, temperature range of the exposure time, and the conductivity of the use dilution. The system provides printed documentation of each cycle. In addition, the Verify System 1E Chemical Indicator is available for routine monitoring of the peracetic acid concentration of the S40 Sterilant use dilution.

SUMMARY OF NONCLINICAL/BENCH PERFORMANCE TESTING

Non-clinical performance data were provided to address the following areas:

Spore Strip Characterization

[1] FDA Guidance Document, Guidance for Industry and FDA Reviewers: Content and Format of Premarket Notification [510(k)] Submissions for Liquid Chemical Sterilants/High Level Disinfectants, released January 3, 2000

Viable spore population pre-processing and post-processing with Builders only:
Testing was conducted to demonstrate that the viable spore population on the Spore Test Strip meets its specification for the starting spore population of at least 1.5 x 10^5 cfu prior to processing and of at least 1.0 x 10^5 cfu following processing through a complete System 1E liquid chemical sterilization cycle under simulated use conditions in the absence of the active ingredient. Samples from 3 lots of Spore Test Strips were processed in the System 1E processor through a complete cycle with Builders only solution (no peracetic acid) or left unprocessed. Note: In the System 1E processor, the active ingredient in the S40 sterilant, peracetic acid (PAA), is combined with inert ingredients (Builders) to form a use dilution. Testing was conducted with the C1220 container/tray, which was modified with the addition of multiple posts attached to the bottom of the container to which the Spore Test Strips were attached with the transfer clips. Thirteen Spore Test Strips were processed in each run. The viable spore population was determined for each sample. Ten of the 13 Spore Test Strips were macerated and then cultured. The other 3 Spore Test Strips were transferred to growth media for comparison. The testing is summarized in Table 2.

Table 2. Summary of Testing for Viable Spore Population

Sample	Processing	Recovery and Culture
10 Spore Test Strips/Lot	No processing	Macerated in Sterile Water for Irrigation (SWFI), cultured 48 hrs, then enumerated
10 Spore Test Strips/Lot	Processing in System1E with Builders Only cup	Macerated in SWFI, cultured 48 hrs, then enumerated
3 Spore Test Strips/Lot	Processing in System 1E with Builders Only cup	Transferred to growth media and cultured 7 days, then enumerated

The acceptance criteria were as follows:

1. The averaged recovered post-processing population will be ≥ 1.0 x 10^5 cfu/Spore Test Strip.
2. The calculated initial population of each sample (average cfu) must be ± 50% of the calculated mean sample population.
3. The recovered average population of unprocessed Spore Test Strips will be within -50% to +300% of the manufacturer's stated population.

The results are shown in Table 3. All Spore Test Strips showed growth. Incubation of the Media alone showed no growth through 7 days of incubation. During the processing cycle, the PAA concentration was measured at the 3 min exposure time and recorded.

Table 3. Viable Spore Population

Lot		Manu. Stated Population	Recovered Initial Population	% Difference	Recovered Population after Processing
1	Average	1.8 x 10^5 cfu	1.8 x 10^5 cfu	0.0	1.4 x 10^5 cfu
	Range	NA	1.5-2.2 x 10^5cfu	--	1.1-1.7 x 10^5cfu

Lot		Manu. Stated Population	Recovered Initial Population	% Difference	Recovered Population after Processing
2	Average	2.2 x 10^5 cfu	2.6 x 10^5 cfu	-18.2%	1.8 x 10^5 cfu
	Range	NA	2.3-3.0 x 10^5cfu	--	1.4-2.4 x 10^5cfu
3	Average	2.1 x 10^5 cfu	2.3 x 10^5 cfu	-9.5%	1.4 x 10^5 cfu
	Range	NA	1.9-3.1 x 10^5cfu	--	1.1-1.9 x 10^5cfu

NA – Not Applicable

The initial population of all Spore Test Strip samples tested was $\geq$1.5 x 10^5 cfu. The population on each of the Spore Test Strip samples after processing was $\geq$1.0 x 10^5 cfu.

The firm also evaluated the compatibility of the strip material with the S40 Sterilant. Ten Spore Test Strips were exposed to two 6 minute cycles in the System 1E processor with the S40 Sterilant with the PAA concentration at 1845-2374 ppm. The results showed no visible signs of damage to the strip material. In addition, as described below, the strip material was shown not to inhibit growth of the *B. stearothermophilus* spores.

Viable Spore Population on the Spore Test Strip pre- and post-rinse phase: Testing was conducted to demonstrate that the loss of spores from the Spore Test Strip by physical wash-off occurs prior to the rinse phase of the System 1E liquid chemical sterilization cycle. The firm evaluated the recoverable viable spore population from the Spore Test Strip before and after exposure to the rinse phases in the System 1E processor. Testing was conducted with 10 strips from each of three lots of the Spore Test Strip. The spore population on the test strips was evaluated under the following conditions:

- Unprocessed
- Exposure to a full System 1E sterilization cycle with Builders-only (no PAA)
- Exposure to a the System 1E exposure phase only (no rinses) with Builders-only

The spore population of each strip was determined and then the average population for the lot was calculated by calculating the mean of the 10 individual strip populations. The results showed a statistically significant loss of viable spores from the strip during the Builders only exposure phase. However, the loss of viable spores from the strip during the subsequent rinse phases was not statistically significant.

Characterization of the spores on the test strip: Testing was conducted to compare the characteristics of the spores on the test strip with the spores remaining on the test strip following processing in the System 1E with Builders-only.

- The spores on the strip were evaluated by Scanning Electron Microscopy. The micrographs showed that the spores are imbedded in the matrix of the strip and do not sit on the surface of the strip. The spores do not physically attach themselves to the matrix.

- The kill kinetic properties of the Spore Test Strips were evaluated before and after

exposure to the Builders-only cycle. A total of 30 strips were exposed to the full Builders-only cycle and then evaluated for kill kinetics using a beaker of the S40 Sterilant use dilution in a 43°C water bath. A single strip was removed at each of 6 time points (40-140 sec) and immediately neutralized. Unexposed Strips were similarly evaluated. The results were the same for both sets of Spore Test Strips. The results demonstrate that the spores maintain the same kill kinetic properties whether or not they are exposed to the Steris System 1E processor.

Growth inhibition: The strip material used for the Spore Test Strip was evaluated for its potential to inhibit growth of the organism, *G. stearothermophilus*, either before processing or after processing in the System 1E processor with the S40 Sterilant. Testing after processing is intended to demonstrate that the strip does not retain significant levels of the Sterilant that may hinder the outgrowth of the organism. Testing was conducted with three different lots of media. Six strips were autoclaved and then clipped in a tray in the System 1E processor and exposed to a full cycle with the S40 Sterilant (2004-2045 ppm PAA, pH 6.35-6.72) and then transferred to growth media. 2 strips were placed in each vial of media. Six additional strips, which had also been autoclaved, but were not processed through the System 1E processor were similarly transferred to growth media. After 2 hrs incubation, each tube was inoculated with an average of 18.7 cfu *G. stearothermophilus* and then incubated at 55-60°C for 7 days. The vials were evaluated daily for growth. All vials showed growth within 24 hrs of incubation. The results demonstrate that the strip material does not inhibit growth, either before or after processing in the System 1E processor. Positive and negative controls showed that the growth media can support growth of low numbers of organisms and that the strip and the media were sterile prior to use.

Because the transfer clip contacts the Spore Test Strip, the firm evaluated the growth inhibition potential of the transfer clip following exposure in the System 1E processor with the S40 sterilant use dilution. The processed clip was placed in growth media and then inoculated with low numbers of *G. stearothermophilus* spores and examined for growth. All samples showed growth within 24 hrs. The results show that the clip does not have inhibitory effects on the *G. stearothermophilus* outgrowth.

Media Validation: Testing was conducted to demonstrate that both the solid and liquid culture media used in the performance studies are capable of supporting growth of a low number of test organisms. Testing was conducted under the following conditions:

- 10% variation in volume
- 10% variation in pH indicator
- Presence or absence of pH indicator

The results show that both the solid and liquid culture media support growth of low numbers of test organisms under the above conditions.

Effectiveness of Neutralization Method: Testing was conducted to demonstrate ability of the method of neutralization (dilution with 0.048% sodium thiosulfate) to effectively

neutralize the PAA and not adversely affect the growth of the organisms. Testing was conducted under the following conditions:

Sodium thiosulfate with Spore Test Strip - strip exposed to S40 Sterilant
- 10 vials inoculated with approximately 10^3 cfu/ml
- 1 vial not inoculated (Negative control)

Sodium thiosulfate with Spore Test Strip - strip exposed to deionized water
- 10 vials inoculated with approximately 10^3 cfu/ml
- 1 vial not inoculated (Negative control)

Sodium thiosulfate with Spore Test Strip - strip exposed to S40 Sterilant
- 1 vial not inoculated – test strip used to check for presence of detectable PAA – the results were negative

Sterile deionized water with Spore Test Strip
- 10 vials inoculated with approximately 10^3 cfu/ml
- 1 vial not inoculated (Negative control)

The PAA concentration was 1487-1588 ppm over the run. The results show equivalent growth in all inoculated tubes. The study validates the neutralization method.

Stability of Color

Testing was conducted to demonstrate that the culture medium maintains a stable color or color change following growth for a minimum of 7 days at 55-60°C. A color change from red to yellow during incubation indicates microbial growth. Three lots of Spore Test Strips and 30 vials of media from each of 3 lots of media were evaluated. The samples were incubated at 55-60°C and monitored daily for growth or color change for 7 days.

- 10 vials – negative control; no inoculation
- 10 vials – inoculated
- 10 vials – inoculated with <100 cfu *G. stearothermophilus*

The results indicate that all inoculated vials show growth and color change to yellow within 24 hrs with no reversion for the duration of the 7-day incubation. All of the un-inoculated vials show no growth and no color change over the same period of time and conditions.

Reduced Incubation Time

Testing to support a reduced incubation time (RIT) of 24 hrs for the Spore Test Strips following exposure to the S40 Sterilant was conducted per the FDA recommended protocol. Testing was conducted with newly manufactured Spore Test Strips and with Spore Test Strips at intervals following storage of 3 months, 6 months, 9 months, and 12 months. The Acceptance Criteria were:

- 30-80% Spore Test Strips show growth
- ≥97.0% growth correspondence with 7 day growth per the FDA guidance

For the 0 and 3 month time points, the samples were exposed to the sterilant in the Use Dilution Vessel at 43°C and about 1635 ppm PAA. Media color was recorded at 18, 24, 48, 72, 96, 120, 144, and 168 hours. One hundred Spore Test Strips were tested per lot per time point. The exposure cycle times varied per lot of Spore Test Strip. The test results in the Use Dilution Vessel for the 0 month and 3 month samples are summarized in Table 4.

Table 4. Reduced Incubation Time of Strips Exposed in the Use Dilution Vessel

Lot	Time Interval	Incubation Time		% Growth after 18 hrs	% Growth after 7 days	% Growth Correspondence
		18 hrs	7 days			
1	0 mos	65/100	65/100	65%	65%	100%
	3 mos	46/100	47/100	46%	47%	97.8%
2	0 mos	47/100	48/100	47%	48%	97.9%
	3 mos	45/100	45/100	45%	45%	100%
3	0 mos	54/100	54/100	54%	54%	100%
	3 mos	49/100	49/100	49%	49%	100%

Temperature and pH were recorded prior to addition of PAA, after the addition of PAA, and then again at the end of the run. PAA concentration was recorded after the addition of PAA and then again at the end of the run. The data indicate that the PAA concentration range for the 100 runs was 1454-1712 ppm after the addition of PAA and 1348-1579 ppm at the end of the run. The results of the RIT testing support reduced incubation times of 18 hours and 24 hours.

For the 6 month, 9 month, and 12 month time points, the samples were exposed to the S40 sterilant in the System 1E processor. For testing in the System 1E processor, the C1140 container/tray was modified with the addition of a stainless steel rod attached across the tray. This configuration was intended to allow 10 Spore Test Strips to experience sterilant contact for an equivalent time period. The tray is kidney shaped and fits snugly into the well of the square tray that fits into the chamber of the System 1E processor chamber. One hundred Spore Test Strips from each of 3 lots and 100 vials of media from 3 lots were used for testing in the System 1E processor with the S40 Sterilant cup using fractional cycles to obtain 30-80% growth. Ten Spore Test Strips were clipped to the stainless steel rod that is attached across the C1140 tray for each run. The cycle was aborted to achieve the partial cycle times. The retrieved Spore Test Strips were incubated at 56±2°C. The test results are summarized in Table 5. The results of the RIT testing support a reduced incubation time of 18 hours and 24 hours over 12 months of storage.

Table 5. Reduced Incubation Time of Spore Strips Exposed in the System 1E

Lot	Time	Incubation Time	% Growth	% Growth	% Growth

	Interval	18 hrs	7 days	after 18 hrs	after 7 days	Correspondence
1	6 mos	74/100	74/100	74%	74%	100%
	9 mos	71/100	71/100	71%	71%	100%
	12 mos	69/100	69/100	69%	69%	100%
2	6 mos	73/100	73/100	73%	73%	100%
	9 mos	50/100	50/100	50%	50%	100%
	12 mos	62/100	62/100	62%	62%	100%
3	6 mos	63/100	63/100	63%	63%	100%
	9 mos	68/100	68/100	68%	68%	100%
	12 mos	76/100	76/100	76%	76%	100%

Spore Resistance Characteristics

Testing was conducted to determine the resistance characteristics of the spores on the Spore Test Strip to the S40 Sterilant use dilution using a "Use Dilution Vessel" under static conditions below the minimum recommended concentration (MRC) of 1800 ppm PAA. The firm conducted the following tests with the Spore Test Strip in a Use Dilution Vessel:

- D-value characterization
- Survival/Kill Window
- Reduced Incubation Time validation testing (see section above)

The Spore Test Strips used in these studies were manufactured from three different *G. stearothermophilus* spore crops. Three different media lots also were used. One of the media lots was replaced with a fourth lot at the 6 month interval because of an insufficient supply of the original lot remaining to complete the shelf life study. The original media lot had been damaged by a water leak into the interior storage compartment of the environmental chamber which soaked the media samples and containers. Numerous vials were soaked with water or showed signs of mold growth. Therefore, approximately half of the vials put up for the shelf life study were removed and disposed. The firm decontaminated the chamber. Inconsistent results were obtained with the remaining media, which was subsequently replaced with the backup lot, which had been stored under the similar conditions. Lots were of similar age.

The Use Dilution Vessel consists of a 150 ml sterile beaker and a sterile stir bar in a water bath held at 43 ± 2°C. The solution temperature was below the minimum in the System 1E of 46°C. Prewarmed Builders solution (100 ml) was added and then a volume of S40 Sterilant concentrate was added to achieve the desired final PAA concentration. The Spore Test Strip was exposed to the PAA in the Use Dilution Vessel , during which the PAA concentration and pH were determined. The Spore Test Strip was removed from the vessel just prior to the full 6 minute exposure and then neutralized in sodium thiosulfate. The Spore Test Strip was then cultured. The Use Dilution Vessel reduces the significant kill that occurs during the fill and warm/mix phases of processing in the Steris System1E processor. It is intended to minimize loss of spores from the test strip due to wash-off and thus permit determination of resistance for the Spore Test Strip to the S40

Sterilant use dilution. An ANOVA analysis of the mean Spore Test Strip population with exposure to the Builders only and with no exposure shows that there is no statistical difference between populations of unexposed Spore Test Strips and those exposed in the Use Dilution Vessel. Eight different tests were run to show the reproducibility and consistency of the PAA concentration and pH in the Vessel during the 6 minute exposure period.

D-value characterization: The D-value specification for the System 1E Spore Test Strip was originally set at ≥ 15 seconds. A survivor curve was constructed using a PAA concentration of 1635 ppm, which is below the MRC of 1800 ppm PAA for the 6 minute cycle in the System 1E processor. The curve shows linearity over 70 seconds. Therefore, the Holocomb-Spearman-Karber (HSK) calculation was used to determine the D-value. Using this method, the D-value is determined as the negative reciprocal of the slope of the survival curve. In comparison, the Stumbo-Murphy-Cochran (SMC) method uses a single time point for calculation of the D-value.

Three lots of Spore Test Strips were used in the study. Five Spore Test Strips were placed in the Use Dilution Vessel with a PAA concentration of about 1635 ppm. Temperature and pH were recorded prior to addition of PAA and temperature, pH and PAA concentration were recorded after the addition of PAA and then again at the end of the run. The data indicate that the PAA concentration range for the twenty runs was 1459-1712 ppm after the addition of PAA and 1413-1551 ppm at the end of the run. One Spore Test Strip was retrieved at each time point as follows: 41 seconds – all survive; 92 seconds and 98 seconds – mixed survival/kill; and 225 seconds and 300 seconds – all killed. Following exposure, the Spore Test Strips were placed in neutralizer and then transferred to growth media and incubated at 55-60°C for 7 days. Samples were scored daily for growth. 20 Spore Test Strips were evaluated per lot per time point for a total of 100 Spore Test Strips evaluated per lot. The results of the study are shown in Table 6 along with a comparison to the D-value determined using the SMC method at 2 different quantal times. All D-values meet the initial specification for the System 1E Spore Test Strip of ≥ 15 seconds.

Table 6. D-value Determination – Comparison of Two methods

Lot	HSK	SMC	
		1.53 min	1.63 min
1	23 sec	18 sec	18 sec
2	24 sec	19 sec	18 sec
3	18 sec	16 sec	17 sec

The D-values was calculated for each lot of S40 Sterilant use dilution and then averaged to establish a mean D-value of 19 sec with a standard deviation of 2.4 sec. The limits of the D-value were set using the following formula: Mean D-value ± 3 x standard deviation. Therefore, the D-value limits were established as 12-26 sec.

Survival/Kill Window: For the survival/kill study, 2 Spore Test Strips were placed in the Use Dilution Vessel with a PAA concentration of about 1635 ppm. One Spore Test Strip was removed at 41 seconds and the other was removed at 225 seconds. The results of

testing with 3 lots of Spore Test Strips are in Table 7. Temperature and pH were recorded prior to addition of PAA; temperature, pH and PAA concentration were recorded after the addition of PAA and then again at the end of the run. The data indicate that the PAA concentration range for the 50 runs was 1445-1712 ppm after the addition of PAA and 1348-1579 ppm at the end of the run. A total of 100 Spore Test Strips were evaluated per lot. Samples were scored daily for growth.

Table 7. Survival/Kill Results

Spore Test Strip Lot	# Growers/# Tested	
	41 sec	225 sec
1	50/50	0/50
2	50/50	0/50
3	50/50	0/50

The survival and kill times were calculated based on the specifications for a manufacturing population of 1.5×10^5 cfu and the D-value limits. The specifications were calculated as all survive at 38 seconds and all kill at 239 seconds when tested at 1635 ppm PAA using the following formulas:

Survival Time = [log (minimum population) -2] x minimum D-value
= [log (1.5×10^5) – 2] x 12 sec
= 38 sec

Kill Time = [log (minimum population) + 4] x maximum D-value
= [log (1.5×10^5) + 4] x 26 sec
= 239 sec

SIMULATED USE TESTING IN STERIS SYSTEM 1E

Simulated use testing with the Spore Test Strips was conducted in the System 1E processor under pass and fail conditions to demonstrate the expected performance of the Spore Test Strips. Testing in the System 1E processor was conducted without use of an instrument load in the final finished System 1E processor. The empty load represents worst case conditions because, with no load present, the sterilant concentrate is diluted to its minimum PAA concentration. Testing was conducted with the C1220 container/tray, which was modified with the addition of multiple posts attached to the bottom of the container to which the Spore Test Strips were attached with the transfer clips. This container/tray was used for post processing the builders-only population testing, in-processor testing and growth inhibition testing. The shape of the tray is a long oval and fits snugly into the well of the square tray that fits into the System 1E processor chamber.

Eighty Spore Test Strips per lot were evaluated for growth following exposure to a Pass cycle with the S40 Sterilant cup (at >1800 ppm PAA) and following exposure to a cycle with a Builders only cup (no PAA). The Spore Test Strips were clipped to posts in a modified 1220 tray. Ten Spore Test Strips were processed in each run in the System 1E processor. The PAA concentration and pH were measured at 3 minutes after the start of the exposure phase. The PAA concentration was 1995-2410 ppm during processing of

the Spore Test Strips in the Pass cycle with the Sterilant. Prior to running the test cycles, the transfer clips were processed through a sterilization cycle in the System 1E with the Sterilant. The transfer clips were then used to attach the Spore Test Strips to the posts in the tray and then to transfer the Spore Test Strips to the growth media. In the results, 0/80 Spore Test Strips show growth following exposure in the Pass cycle and 80/80 Spore Test Strips show growth following exposure to the Builders Only cycle with a 6 minute exposure phase. The Spore Test Strips performed as expected under pass and fail conditions in the System 1E liquid chemical sterilization cycle.

SHELF LIFE/STABILITY

The subject device is not sterile and is not reusable. Shelf life/stability testing was conducted to support a shelf life of 12 months when stored at 2-24°C and 30-80% RH away from sterilizing agents and excessive heat. Three lots of Spore Test Strips from 3 different lots of *G. stearothermophilus* and 3 lots of media were evaluated in the study. The Spore Test Strips and media were stored at 24±2°C at 80%±10% RH. Samples were tested at 3 month intervals – 0, 3, 6, 9, and 12 months. Spore Test Strips and media were randomly selected, although Spore Test Strips and media were paired up before storage. Samples were evaluated, as shown below, according to the criteria described for the performance and characterization studies.

a. Viable spore population pre and post processing with builders only (10 Spore Test Strips per lot)
b. D-value characterization study (10 Spore Test Strips per lot per 5 exposure times)
c. Survival/Kill window (50 Spore Test Strips per lot per survival time or kill time)
d. Performance in the System 1E (100 Spore Test Strips per lot)
e. Stability of Read

In addition, Reduced Incubation Time was assessed for Spore Test Strips exposed in the Use Dilution Vessel and for Spore Test Strips processed in the System 1E processor. See Reduced Incubation Time section above.

As shown in Table 8, all Spore Test Strips passed the test criteria to support a shelf life of 12 months.

a. **Viable spore population pre and post processing with builders only (no PAA):** Test samples were evaluated in the 1220 tray. The acceptance criteria were:
    - Average recovered post processing population is $\geq 1.0 \times 10^5$ cfu/Spore Test Strip
    - The strips must not show any signs of damage after being exposed to two consecutive liquid chemical sterilization cycles
    - Calculated initial population of each sample must be ±50% of the calculated mean sample population
    - Average recovered population will be -50% to +300% of stated manufacturing population

Table 8. Stability of Viable Spore Population

Lot	Time Interval	Manu. Stated Population	Recovered Initial Population	Recovered Population Post Processing
1	0 mo	1.8×10^5 cfu/Strip	1.8×10^5 cfu/Strip	1.4×10^5 cfu/Strip
	3 mo		1.7×10^5 cfu/Strip	1.1×10^5 cfu/Strip
	6 mo		1.1×10^5 cfu/Strip	8.3×10^4 cfu/Strip*
	9 mo		1.6×10^5 cfu/Strip	1.3×10^5 cfu/Strip
	12 mo		2.1×10^5 cfu/Strip	1.1×10^5 cfu/Strip
2	0 mo	2.2×10^5 cfu/Strip	2.6×10^5 cfu/Strip	1.8×10^5 cfu/Strip
	3 mo		1.9×10^5 cfu/Strip	1.3×10^5 cfu/Strip
	6 mo		2.3×10^5cfu/Strip	2.1×10^5cfu/Strip
	9 mo		1.8×10^5 cfu/Strip	1.4×10^5 cfu/Strip
	12 mo		2.5×10^5cfu/Strip	2.0×10^5cfu/Strip
3	0 mo	2.1×10^5 cfu/Strip	2.3×10^5 cfu/Strip	1.4×10^5 cfu/Strip
	3 mo		1.9×10^5 cfu/Strip	1.8×10^5 cfu/Strip
	6 mo		2.3×10^5cfu/Strip	1.8×10^5 cfu/Strip
	9 mo		1.9×10^5 cfu/Strip	1.7×10^5 cfu/Strip
	12 mo		1.6×10^5cfu/Strip	1.5×10^5 cfu/Strip

The data for lot 1 at the 6 month time point* did not meet the specifications. The petitioner explained that the agar used in the population assay was too hot when used and concluded that because the results at the 9 and 12 month time points met specifications, the low populations at the 6 month time point were artificially low due to loss in viable spores on the strips because the agar was too hot. With this explanation, it was shown that all three lots of the Spore Test Strip meet the acceptance criteria after 12 months under the label storage conditions.

b. **D-value determinations by fraction negative method:** PAA concentration was ≤1800 ppm and the temperature was 43±2°C for all runs. The acceptance criteria were:

- D-value: 12-26 min
- ±20% of initial calculated D-value for Spore Test Strip lot

The results are summarized in Table 9.

Table 9. D-Value Determined by Fraction Negative Method

Lot	Time Interval	D-value	% Difference from Initial Value
1	0 mo	23.0 sec	NA
	3 mo	21.1 sec	-8.3%
	6 mo	21.3 sec	-7.4%
	9 mo	17.6 sec	-23.6%
	12 mo	19.8 sec	-13.9%
2	0 mo	23.5 sec	NA
	3 mo	18.9 sec	-19.6%
	6 mo	19.5 sec	-17.0%
	9 mo	19.6 sec	-16.6%

Lot	Time Interval	D-value	% Difference from Initial Value
	12 mo	15.6 sec	**-33.6%***
	15 mo	21.1 sec	-10.3%
3	0 mo	18.0 sec	NA
	3 mo	19.5 sec	8.3%
	6 mo	20.8 sec	15.6%
	9 mo	20.8 sec	15.6%
	12 mo	17.0 sec	-5.6%

* Extra point added to evaluate whether D-value had really dropped

During incubation, one heating block was at 24°C at the 12 mo time point for lot 2.* The samples were transferred to another heating block at the proper temperature and incubated for an time point of 15 mo. The results did not change and that there was no negative impact on the study.

c. **Survival/Kill Times:** Spore Test Strip samples were exposed in the System 1E processor to a PAA concentration of <1800 ppm. The results showed all samples exposed for 41 seconds showed growth and all samples exposed for 225 sec showed no growth. The acceptance criteria were:

- All survive: 38 sec
- All kill: 239 sec

Although the specifications are 38 seconds survival time and 239 seconds kill time, testing was conducted at a longer survival time and a shorter kill time.

d. **Processor Performance:** Testing was conducted with the C1220 tray in the System 1E processor as previously described. The firm indicates that the PAA concentration was >1800 ppm for these cycles. The acceptance criteria were:

- All Spore Test Strips processed with S40 Sterilant Concentrate must not show growth for 7 days of incubation time.
- All Spore Test Strips exposed to S40 Builders only should show growth within 7 days incubation time.
- All in situ tests must complete a full cycle.

All Spore Test Strips processed with the Sterilant were successfully killed at each stability time point and all Spore Test Strips processed with the Builders only (no PAA) showed growth within 1 day of incubation.

e. **Stability of Media Color:** Testing was conducted to assess the stability of the media color change and turbidity. The samples were observed daily for color. The Acceptance Criteria were:

- All inoculated samples demonstrate growth by color change and maintenance

of that color change
- All negative controls should maintain the same color for the 7 days of incubation

All media samples inoculated with a Spore Test Strip or suspension indicated growth within 24 hrs of incubation at 55-60°C. The color change was maintained throughout the remainder of the incubation period. All negative controls maintained the red color.

BIOCOMPATIBILITY/MATERIALS

The Verify Spore Test Strip for S40 does not come in contact with patients or with devices that come in contact with patients. The Indicators are handled by healthcare personnel with gloves.

LABELING

The de novo petition contains appropriate instructions for use, examples of the Certificates of Analysis, which identifies the viable spore population, D-value, and survival and kill times for the lot of spore test strips, and labels for the device. The instructions for use include the indications for use, performance characteristics, use instructions and interpretation of the results, storage conditions, expiration date, performance limitations, appropriate caution statements, and disposal instructions in accordance with 21 CFR 807.87(e). The information provided is acceptable.

RISKS TO HEALTH

Table 10, below, identifies the risks that may be associated with use of the Spore Test Strip and the measures recommended to mitigate these risks.

Table 10. Risks to Health and Recommended Mitigation Measures

Identified Risk	Recommended Mitigation Measures
User handling error due to false fail Spore Test Strip device result due to technical malfunction	Spore Strip Characterization Simulated Use Testing Shelf Life Labeling
User handling error due to false pass Spore Test Strip device result due to technical malfunction	Spore Strip Characterization Simulated Use Testing Shelf Life Labeling

Identified Risk	Recommended Mitigation Measures
User handling error due to misunderstanding Spore Test Strip device use instructions	Labeling

SPECIAL CONTROLS:

In combination with the general controls of the FD&C Act, the Steris Verify Spore Test Strip for S40 for use in the Steris System 1E Liquid Chemical Sterilant Processing System is subject to the following special controls to provide reasonable assurance of the safety and effectiveness of the device type of Spore Test Strip:

(1) Spore Strip Characterization
   i. Population of viable spores on strip shall be a minimum of 5 $\log_{10}$ after physical wash-off of spores from the strip by exposure to liquid chemical sterilant in the liquid chemical sterilant processing system, which should be validated over the claimed shelf life.
   ii. The resistance characteristics of the viable spores on the strip should be defined and be validated over the claimed shelf life.
   iii. The Spore Strip description should address the carrier material, how the spores are placed on the carrier, and whether there is any feature that minimizes spore wash-off. Bacteriostasis of the Spore Strip materials should be evaluated.
   iv. Incubation time for viable spores on the strip should be validated under the specified incubation conditions over the claimed shelf life.

(2) Simulated Use Testing: Simulated use testing should demonstrate performance of spore test strip in liquid chemical sterilant/high level disinfectant under worst case in use conditions over the claimed shelf life.

(3) Labeling: Labeling should specify appropriate instructions, warnings, cautions, limitations, and information relating to viable spore population, resistance characteristics, and interpretation of a "no growth" result.

CONCLUSION

After review of the information submitted in the de novo petition and through interaction with the petitioner, the Steris Verify Spore Test Strip for S40 indicated to provide users with a means to assess spore kill by a liquid chemical sterilant in a liquid chemical sterilant processing system, can be classified in class II with the establishment of the special controls as outlined above.

The special controls appropriately mitigate the identified risks of the Steris Verify Spore Test Strip for S40 and provide reasonable assurance of the safety and effectiveness of the device. Like most class II medical devices, spore test strips used in a healthcare environment are subject to the Quality Systems regulation (QS regulation) as set forth at 21 CFR Part 820. Spore test strip device manufacturers must also comply with the requirements of the Medical Device Reporting (MDR) regulation 21 CFR Part 803.

The de novo petition for the Steris Verify Spore Test Strip for S40 is granted and the device is classified under the following:

Product Code: OVY
Device/Product Name: Spore Test Strip
Class: II
Regulation: 21 CFR 880.6887