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CQV #5: Problems in Aseptic Technique Qualification

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CQV – Compliance in Quality and Validation – presents real-life stories reflecting compliance problems in the pharmaceutical, medical device, and related industries. Previous discussions have included:

> *CQV #1: Overview and Invitation to Participate, published in Journal of GXP Compliance (JGXP), Volume 22, #5, and in Journal of Validation Technology (JVT), Volume 24, #5*
>
> *CQV #2: Like-for-Like Problems. JVT, V24, #3*
>
> *CQV #3: More Life-for-Like Problems. JVT, V24, #5*
>
> *CQV #4: Animal Tales. JGXP, V22, #6.*

Readers are invited to contribute content, suggestions, comments, and ideas to improve this feature. All published content will be anonymous – there will be no connection to companies or organizations. CQV will be most useful when the quality and validation communities submit experiences that will help colleagues improve actual work situations. Please contact coordinators Paul Pluta atpaul.pluta@comcast.net or Melissa Carella atmelissa.carella@cbinet.com with content, comments, suggestions, or topics for discussion.

INTRODUCTION

This discussion addresses problems experienced with qualification or competency testing of manual aseptic technique performance. This testing verifies acceptable aseptic skills by technicians in their functional job responsibilities. The term “qualification” in this discussion is defined to confirm acceptable technical performance; pharmacy sterile compounding practices as regulated by USP <797> requires verification of technician “competency.” Thus, qualification and competency are synonymous terms in describing aseptic technique testing.

Job activities addressed by this testing encompass IV medication preparation in hospitals, microbial identification testing in pharmaceutical industry microbiology laboratories, manufacturing processes in FDA 503b compounding pharmacies, aseptic research activities in clinical settings or university labs, aseptic processes in medical device manufacturing, and similar activities. Successful test performance confirms the acceptable techniques performed by technicians and approves them for future aseptic work. Testing is then periodically repeated to demonstrate ongoing personnel competence based on the procedural risk level.

There are multiple variations of aseptic testing procedures utilized by organizations depending on their functional applications. Testing may include aseptic transfers of microbial growth medium, multiple aseptic dilutions, sterile filtrations, touch plates, contact testing of surfaces, air sampling, and combinations thereof. Test procedures may be a single test on one day; others may use multiple tests performed on consecutive days.

Discussion Topics

Topics addressed in this discussion include the following:

- Aseptic testing description. A representative aseptic test procedure including supporting activities is described.
- Consequences of test failures. What happens if there is a test failure?
- Test problems. Specific testing problems including invalid practices and test mistakes.
- The role of management. The impact of management in aseptic operations.

Content and problems described are based upon experiences provided by quality managers and individuals from several organizations in industry, pharmacy practice, and academia. We appreciate their input into this discussion.

ASEPTIC TESTING DESCRIPTION

Aseptic technique qualification testing may be simply described: A properly trained technician executes an approved protocol requiring multiple aseptic transfers and contact tests using sterile supplies in a qualified aseptic work area. However, the individual activities required for this testing, both in execution and supporting activities, are numerous and detailed. The qualification protocol should represent the worst-case aseptic activities performed by the technicians at the site. Test samples in correctly prepared media are then incubated under appropriate temperature and time requirements. After incubation, test samples must be in compliance with predetermined requirements; e.g., USP <797>. This qualification procedure confirms aseptic processes rendering products and processes sterile and uncontaminated. All facilities, properly maintained equipment, materials, and approved procedures in the test must be tightly controlled to provide the basis for a valid test. All activities associated with the above should be documented, verifiable, and available for review. For reference documentation supporting test procedures and performance see USP <797> (1-7), USP <71> (6), USP <1116> (7), FDA cGMP's for aseptic processing (8), and E. Kastango on a guide for aseptic technique verification (9), and E. Kastango on the top 10 gaps in USP <797> (10).

Test Supplies

Supplies needed for aseptic technique qualification testing is dependent upon the processes being tested. These may include sterile syringes and needles, sterile empty vials, vials or bags containing sterile growth media, sterile water for injection, sterile Petri dishes for finger-tip touch impression tests, sterile RODAC (Replicate Organism Detection and Counting) plates or convex contact plates for testing work surfaces, sterile filters, sterile filtration equipment, and associated supplies. All ancillary test supplies must be sterile. The exterior surfaces of all of the test items and supplies to be used must also be sanitized before the donning of sterile gloves.

Microbial Growth Medium

The microbial medium used in all tests is a general-purpose medium for growth of representative organisms. Soybean-casein digest (SCD) agar containing lecithin and polysorbate 80 (Tween® 80) is typically used for testing. SCD medium is also known as trypticase soy agar (TSA). Additional growth media can include fluid thioglycolate medium for anaerobes and malt extract agar for yeasts and molds. All media must be properly prepared, sterile, stored properly prior to use, tested to support grown, and with results properly interpreted for the testing; refer to USP <71> Sterility Tests (6).

Aseptic Facility

Depending on the site application, an ISO 5 (Class 100) room or equivalent certified laminar air flow work station (LAFW) is typically used for testing. The environment and facility used for testing must be sterile in support of a valid qualification test.

Test Procedure

After the growth medium is prepared, aseptic tests are performed in a defined order. Key points impacting testing that may be commonly overlooked are listed in each section.

1. Media preparation
2. Aseptic transfer procedure
3. Fingertip touch plates
4. Surface sampling with contact plates
5. Air sampling
6. Incubation
7. Test results, requirements, and interpretation

8. Documentation.

1. Media Preparation. A general multiple purpose non-selective growth media such as SCD or TSA may be used in media-filled bags/vials, Petri dishes/plates and air sampling strips. If prepared in-house, media must be correctly prepared and stored. If obtained commercially, containers must be properly stored.

- Growth media containing lecithin and polysorbate 80 are used in the media formulation.
- Growth media containing antimicrobials, preservatives, or other growth inhibitors must not be used in the media formulation.
- Prepared media is to be stored under refrigeration. Note the time limitations on storage.
- A growth promotion test must be performed prior to technician testing to preclude possible false negative test results.

2. Aseptic Transfer Procedure. The aseptic transfer qualification procedure should be developed to simulate the most challenging worst-case representation of aseptic processes performed at the site. A typical aseptic transfer procedure generally includes the following:

a. Garb appropriately (head cover, gown, gloves, etc.) for the work to be demonstrated
b. Sanitize the work environment per site written procedures
c. If sterile empty vials are not available, using an appropriately sized sterile syringe and needle, aseptically remove all of the water from a vial Sterile Water for Injection or equivalent to provide an empty sterile vial.
d. Aseptically transfer sterile media from media-filled bag or bottle into an empty sterile vial.
e. Using the same syringe and needle, transfer multiple small volumes of sterile medium to demonstrate multiple transfers from bag into vial. For example, USP <797> suggests the transfer of three sets of four 5 mL aliquots of sterile SCD into each of three 30 mL sterile vials for low-risk level CSP's (2).
f. Aseptically transfer multiple small volumes of sterile medium from a vial to a sterile growth medium filled bag to demonstrate multiple transfers from a vial into a bag.
g. Label and incubate.

3. Finger-tip Touch Plates. Touch plate tests, also known as gloved finger-tip tests, are performed immediately after completion of the media-fill transfers but before the cleaning and sanitization of the work area.

- Gloves must not be changed prior to performing touch plate test
- Gloves must not be disinfected immediately before the test
- Media plates must be a room temperature for testing
- A typical procedure comprises light touching and rolling of the fingertips of both hands for 1-2 seconds per finger on the medium surface.
- Close the plate, lightly tape shut, label and incubate.

4. Surface Sampling with Contact Plates. Tests using surface contact plates, also known as RODAC plates, are performed immediately after completion of other tests and prior to the cleaning and sanitization of the work area. The precautions described for the finger-tip touch plates are similarly followed for contact plates.

- A typical contact plate test comprises gentle touching of plate to work test surface. Do not slide the plate side-to-side on the test surface. Close the plate, lightly tape shut, label and incubate.
- Swabbing of the work surfaces followed by sterile medium inoculation may also be utilized if direct surface contact plates are not possible.

5. Air Sampling. Sampling of the work environment air is to be performed throughout the performance of the aseptic testing. Air sampling may be performed via the use of a Reuter centrifugal impaction principle (RCS®) air sampler.

- Settling plates should not to be used as this approach to air sampling is not a discriminating test method and can therefore result in false negatives.

6. Incubation. Incubation must be correctly done to enable growth of contamination. Incubate at correct temperatures and times along with positive and negative controls to confirm test acceptability as per USP <797> (1) and USP <71> (6).

- Incubate plates upside down so that the condensation does not drip onto surface and possibly interfere with microbial growth.
- Observe samples at recommended times for cloudiness in liquid media, and perform counts of colony forming units

(cfu's) on the plates/strips.

7. Test Results, Requirements, and Interpretation. Compile all test results. The test results are to be in compliance with USP <797> requirements.

8. Documentation. All training and qualification documentation including media preparation and growth promotion testing must be properly documented and maintained in a readily retrievable manner ready for inspection.

CONSEQUENCES OF TEST FAILURES

Failure of a technician qualification test is a serious event requiring an investigation and the implementation of corrective/preventive actions (CAPA's). In failures, microbial growth is observed in the aseptic preparations suggesting that the technician has not performed acceptable aseptic technique. Failure casts suspicion on the previous work done by the technician since the previous acceptable aseptic technique testing. In the event of a failure, the technician should not be allowed to perform the preparation of compounded sterile products until a successful investigation, retraining, retesting and/or other CAPA actions are successfully implemented. Investigations should include all previous determinations, products, and test results since the last successful competency testing – a significant undertaking. The consequences of a failed aseptic qualification test can be serious indeed – obviously to the patients but also as well to the organization and to the technician.

TEST PROBLEMS

Several categories of aseptic test problems were described by quality and validation managers.

- Invalid practices. Procedures that are utilized that can inhibit or minimize detection of microbial growth and false negative test results.
- Test mistakes. Errors executing the test procedures that can nullify tests or can possibly result in false negatives.
- Insufficient testing. An organization that does not test all personnel involved in aseptic procedures and/or does not perform all of the tests.
- Inadequate investigations -- Test failures of technician technique or contact contamination are not thoroughly and comprehensively investigated.

Invalid Practices

Quality managers and associated individuals related several practices that could possibly result in the reduction of the qualification challenges which in turn can minimize the risk of microbial test failures. Such practices are due to inadequate training and/or supervision.

The qualification of aseptic technique should be conducted under worst-case challenge conditions that are most likely to demonstrate the potential for microbial contamination. Quality managers commented that testing is sometimes inadvertently conducted under de facto "best-case" rather than worst case conditions. The following are practices that have been utilized by some organizations resulting in inappropriate "best-case" conditions practices.

First Activity of the Day. Aseptic qualification testing should not be scheduled to be the first activity of the day. Testing as the first activity after cleaning of the work area and before any other work has been initiated can result in "best case" conditions with minimal potential for contamination. Qualification of aseptic technique should be conducted after routine work is completed and before area clean-up at the end of the work day. Likewise, the qualification procedure must not be interrupted once the process begins.

Work Area Sanitization. Technicians must not excessively clean the work area before and in between qualification tests – again resulting in "best case" conditions. For example, the work area is cleaned and sanitized immediately before initiating testing and/or sanitized after media transfer testing but before touch plate and surface contact testing; touch plates or other testing are then immediately performed.

Glove Changing and Sanitization. The donning of new sterile gloves or spraying one's gloves with sterile 70% isopropanol immediately prior to a finger-tip touch test does not challenge aseptic technique, does not represent actual work performance, and therefore must not be done. Changing or sanitizing one's gloves immediately prior to the finger-tip touch test does not represent a worst-case challenge and will consequently bias the results.

Supervision. The person being tested must perform the aseptic procedure on an independent basis. A trained person should

not be coaching and directing the technician on how to perform each step in the qualification process and prompt specific actions. This coaching approach does not demonstrate that the employee can independently perform aseptic procedures. The technician must first be thoroughly trained in aseptic technique prior to undergoing the qualification procedures.

Incubation. Incubation temperatures and times must be strictly followed for the best observation of potential growth. Incubation times must not be shortened which can preclude the detection of slow-growing organisms.

Growth Medium Preparation. Correct preparation of growth media is mandatory. Here are some examples of incorrect practices:

- The use of media that contain the incorrect ingredients is an error. That is, media are used that contain substances that can inhibit the desired growth. For example, phenol in a growth medium can result in an inhibition or the false lowering of the true microbial count.
- Another example is skipping of ingredients in media preparation despite documents recommending their addition. Media must contain all ingredients as specified in the procedures. For example, lecithin and polysorbate surfactants are vital ingredients in media. Quaternary ammonia compounds are neutralized by lecithin while phenolic disinfectants and hexachlorophene are neutralized by polysorbate 80. Together lecithin and polysorbate 80 will neutralize any surface disinfectants such as alcohols.
- An additional example of an incorrect technique is if growth promotion tests are omitted. If growth promotion tests are not conducted to assure that the growth medium will support growth, then false negative results may result if the media cannot support growth.

Standards. Standards/acceptance criteria are sometimes incorrectly set too high, such 5 colony forming units (cfu's), or higher, results for finger-tip touch plates.

- Acceptance criteria that are too lenient are meaningless and are also not in compliance with USP <797>.

Results. Failing test results are mistakenly considered to be invalid where the qualification tests are then repeated.

- Failing results should be documented. Technicians that fail the qualification tests should not be allowed to continue aseptic processing until successful requalification has taken place.
- Inaccurate documentation can also damage the integrity of the site's data and quality program.
- As some individual tests by themselves may not be determinant of successful qualification of aseptic technique, such as only examining the medium filled bag for growth, the qualification of aseptic technique needs to examined on a holistic basis taking all of the separate qualification tests together into account.

Test Mistakes

Additional practices in the testing or supporting activities are sometimes not correctly performed. These errors can prevent actual contamination from being detected. These practices may also be "institutionalized" in procedures as in – "This is the way we do things" – and are therefore not perceived as mistakes.

- Medium is not properly stored between receipt and use in testing. Medium should be stored under refrigeration and visually examined prior to use. Medium that is cloudy or turbid may already be contaminated. Medium that is old, dried, shrunken, cracked, discolored or displaying other obvious signed of deterioration must not be used for testing. New growth promotion tests should be conducted if medium acceptability is questionable and/or if the medium is past its use date.
- Refrigerated stored medium must be allowed to warm to room temperature for testing.
- Growth promotion tests are not performed on medium prepared in-house or on purchased media. There is no assurance that any contamination will actually grow.
- Test plates are tightly sealed for incubation. This prevents aerobic organism growth due to lack of oxygen.
- Contact test plates are stored upright for incubation. This enables condensation to form on medium surface and can potentially interfere with organism growth.
- Medium is incubated at wrong temperature, or incubator temperature controls are not calibrated/certified and as such are unreliable to maintain a constant temperature.
- Incubation is terminated prematurely; slow-growth organisms are thus not able to be observed.

Insufficient Testing

Insufficient qualification testing comprises multiple situations which can result in erroneous results or conclusions. For example, testing only the “best” technicians, i.e., not all personnel who perform aseptic activities are tested which can result in the wrong conclusion that all aseptic processes and people are qualified.

- The site quality program supervisor works during daytime hours and as such aseptic testing can only be conducted during daytime hours only. Technicians who perform the aseptic operations that work during second or third shift are mistakenly never tested since the quality person is not available.
- Part-time technicians, students, interns and/or residents are sometimes not tested or insufficiently trained and tested.
- During qualification, technicians may conduct the media transfers using different techniques compared to their regular work processes. For example, a technician may use a new sterile needle and/or syringe for each transfer in their aseptic technique qualification, but may reuse the same needle and syringe when performing routine transfers. Changing needles and/or syringes between each transfer should not be done during the qualification of aseptic technique.
- An organization may require only one transfer into the media filled bag or vial and then incubate. In this case, one transfer into the media filled container is inadequate as an insufficient number of transfers were done, combined with such transfers into a media filled container is inherently not discriminating as test failures are rarely encountered. Requiring only minimal testing such as only relatively simple media transfers rather than “worst-case” aseptic processes, can result in mistaken/invalid conclusions on the qualification of the technician’s aseptic processes.
- Worst-case challenges are not required to be simulated in competency testing. For example:
    - The preparation of solutions for a penicillin desensitization protocol could entail ~20 dilutions and such transfers are not tested in qualification testing.
    - Non-sterile to sterile high-risk compounding in which bulk non-sterile drug is a starting material requiring sterilizing filtration is not tested in competency testing when indeed such high-risk CSP’s are prepared at the site.
- Transfer is conducted using a sterile marketed drug product solution rather than media which can bias the test to a negative growth outcome.
- Media transfers may be done, but finger-tip touch plates, surface, and/or air sampling tests are not done.

Inadequate Investigations

Test failures indicating substandard technician aseptic technique is a serious problem. Test failures may occur for a multitude of reasons: Insufficient hand hygiene, contamination during garbing, inadequate hand disinfection, inadequate work area disinfection, errors in aseptic technique, etc. These and all failures must be thoroughly investigated and documented. Investigations are inadequate where the root cause(s) of the failure(s) have not been identified and/or corrective actions have not been successfully implemented.

Technicians who fail the qualification testing must not be allowed to continue aseptic activities. Training and retesting must be also be conducted and documented before regular work is allowed to continue.

Test results need to be compiled and trended as a function of time and the technician. Further investigations are indicated upon any detected out-of-trend results. Continued monitoring is necessary to verify that any changes have been successfully implemented (10).

THE ROLE OF MANAGEMENT

While this discussion has focused primarily on errors and mistakes associated with the activities and testing of technicians who perform aseptic techniques, its scope encompasses much more than the individual identified problems. Why do these problems occur? More specifically, does management have influence on the incidence of these problems?

Priority and Support

The attitudes and performance of employees in any organization are directly related to the attitudes of their management. Employees may naturally take “short-cuts” in routine procedures, become complacent, or otherwise jeopardize the quality of the activities they perform under excessive stressful workloads or other difficult working conditions. Due to the scope, complexity, workload, and priorities of organization activities, aseptic activities may easily become “taken for granted”, and therefore “out of sight out of mind.” Then management interest consequently occurs only when there is a problem. The

performance of management who lack interest or are invisible to aseptic work functions can sometimes be interpreted as being insincere in compliance to aseptic performance standards. Managers and responsible individuals in aseptic areas must be visibly supportive of their employees and continually on guard against these normal human tendencies that are especially critical in aseptic performance.

Training

Lack of headcount and/or budgetary constraints can negatively impact the implementation, performance, or expansion of quality systems and programs. Training is often minimized or eliminated in these situations. Several categories of aseptic test problems were identified above including invalid practices and test mistakes that were subtle modifications of best practices. It is only through strong management commitment to quality performance through ongoing training and supervision that these problems may be avoided. When headcount is insufficient or workload is excessive, mistakes and errors can lead to unsafe practices and patient harm. When headcount is minimal, too few trained supervisors are involved; procedures do not exist or may be too vague, incomplete, or obsolete; in-house training is reduced or cancelled, and training at external seminars is not supported. Many of the mistakes described above may indicate personnel who do not have basic training in aseptic techniques. The supervision of aseptic activities may also be delegated to senior or experienced technicians who are not trained and developed for this role. In this case, they may lack the technical education, continuing professional development, and motivation to take active responsibility for supervising aseptic operations.

Management can establish job performance goals of not having any failures in the qualification of aseptic technique. This message can be incorrectly interpreted to mean the technicians should do whatever was necessary to prevent failures – hence the implementation of erroneous practices as described above. Such goal setting can be risky as it can lay the foundation for short-cuts and mistakes.

Management Leadership and a Culture of Quality

Management must lead and set a positive tone by visibly implementing and supporting a quality systems program in the organization. Mangers can demonstrate their support by creating the environment for success by providing a high priority for aseptic activities, budget, headcount, time, training, assuring proper facilities/instruments/supplies/tools for the process and by personally reviewing the test results, and holding people both responsible and accountable.

Management must diligently strive to improve their systems via continuous monitoring, performance improvement (PI) programs, training and education of the personnel who perform aseptic processes such that everyone understands not only the “how to” but also the “why” – the rationale behind the quality system procedures. These compliance activities need to be evaluated and integrated as a continuum (10).

How to achieve these desired behaviors and expectation levels? One practical way is to modify the rewards systems to achieve the desired outcomes. That is, successful quality control programs, with quantifiable metrics, can be incorporated into a manager’s objectives in their annual performance reviews and bonus programs. Further, a qualified trained person can be made responsible for the independent conduction of internal audits where he/she reports the results directly to the department head and/or senior management.

Management must be unwavering in its commitment to quality and compliance standards in order to maintain a high degree of control for the organization, for risk management, and more importantly, patient safety. Only through organizational dedication can a state of control be realized for safe aseptic compounding (10). More importantly, our patients are dependent upon our good aseptic practices.

REFERENCES

1. USP. <797> Pharmaceutical Compounding – Sterile Preparations. USP-37. pp. 410-453.
2. USP. <797> Pharmaceutical Compounding – Sterile Preparations. Media Fill Test Procedure. USP-37. P 416.
3. USP. <797> Pharmaceutical Compounding – Sterile Preparations. Appendix III. Sample Form for Assessing Hand Hygiene and Garbing Related Practices of Compounding Personnel. USP-37. P. 451.
4. USP. <797> Pharmaceutical Compounding – Sterile Preparations. Appendix IV. Sample Form for Assessing Aseptic Technique and Related Practices of Compounding Personnel. USP-37. P. 452.
5. USP. <797> Pharmaceutical Compounding – Sterile Preparations. Appendix V. Sample Form for Assessing Cleaning and Disinfection Procedures. USP-37. p. 453.
6. USP. <71> Sterility Tests. USP-40. pp. 136-143.

7. USP. <1116> Microbiological Control and Monitoring of Aseptic Processing Environments. USP 36. pp. 784-794.
8. FDA. Guidance for Industry – Sterile Drug Products Produced by Aseptic Processing - Current Good Manufacturing Practice. Sept 2004. Available at: https://www.fda.gov/downloads/Drugs/Guidances/ucm070342.pdf Accessed Nov 4 2018.
9. Kastango, Eric S. A Practical Guide to Aseptic Technique Verification: Policies and Procedures that Meet USP Chapter <797> Requirements. Pharmacy Purchasing and Products Magazine. Vol. 2, No. 2. April 2005. Available at: https://www.pppmag.com/documents/V2N2/Asceptic.pdf Accessed Nov 4 2018.
10. Kastango, Eric S. The Top 10 Gaps in USP Chapter <797> Compliance. Cleanrooms & Compounding. Pharmacy Purchasing and Products Magazine. Vol. 6, No. 2. October 2009. Available at: https://www.pppmag.com/documents/V6N10_cleanrooms/p2_4_5_6_7.pdf Accessed Nov 4 2018.
