

tests must include at least two control materials of different concentrations per day. Standards may be used in lieu of control materials, as long as they are not the same as the materials used to calibrate the test system or establish a cutoff.

In reviewing the CLIA regulations concerning control procedures and QA requirements for molecular amplification procedures, the CLIA discussed appropriate control procedures and QA for genetic testing (September 16, 1998 through September 17, 1998). CLIA recommended that controls for genetic testing should be considered for laboratories in general, including ensuring that adequate controls are in place to minimize contamination. This is especially important when performing molecular amplification procedures. To ensure the control of contamination, we have amended the requirements for facilities, formerly at § 493.1204(a) (now at § 493.1101(a)) to require laboratories to be constructed, arranged, and maintained to minimize contamination of patient specimens, equipment, instruments, reagents, materials, and supplies. A uni-directional workflow must be maintained for molecular amplification procedures not contained in closed systems. This must include physically separate areas for specimen preparation, amplification and product detection and, as applicable, reagent preparation. We believe these measures will decrease the potential for contamination to the extent possible in a clinical laboratory.

*Comment:* Several commenters requested clarification of the control requirements for kit systems used for bacterial and fungal identification. One commenter specifically requested the addition of a provision at § 493.1231, Mycology, that would require the testing of each new shipment of test kits or strips used for organism identification with organisms giving positive and negative reactions for each test before or concurrent with testing of clinical isolates. Another commenter questioned whether these systems would be subject to the requirement described at § 493.1202(c)(4) to test at least two levels of control materials each day of testing.

*Response:* We agree with the commenter that in mycology, or any other subspecialty area of microbiology, new shipments of test kits or strips used for organism identification should be tested with organisms giving positive and negative reactions for each test before or concurrent with initial testing of clinical isolates. This includes identification kits or panels that are

inoculated and read manually, and those that are part of an automated instrument system. We are retaining the requirement formerly at § 493.1218(f)(1) (now at § 493.1256(e)(1)) that laboratories check each batch (prepared in-house), lot number (commercially prepared), shipment of reagents, disks, stains, antisera, and identification systems (systems using two or more substrates and/or reagents) when prepared or opened for positive and negative reactivity. We do not believe additional testing of these systems is needed if they are stored and maintained under appropriate conditions. Further testing is only necessary if labile reagents must be prepared or used each time the kit is used or if specified by the manufacturer.

*Comment:* Several commenters requested clarification of the control requirement at § 493.1218(b)(1) for qualitative tests as applied to microbiology procedures. The commenters asked which of the biochemical tests or media used for microbial identification would be considered qualitative tests.

*Response:* Biochemical tests using specific reagents or growth tests that employ selective or differential media (for example, indole tests, citrate media) that are a part of the total system of identification from culture are not considered qualitative tests in microbiology. Therefore, we are retaining the requirement formerly at § 493.1218(f)(1) (now at § 493.1256(e)(1)) that states laboratories must check each new batch (prepared in-house), lot number (commercially prepared), and shipment when prepared or opened for positive, negative, and graded reactivity, if applicable. Specifically, former § 493.1218(f)(4) (now at § 493.1256(e)(1) and (4)) requires each batch of media to be checked before or concurrent with initial use for sterility, and its ability to support, select, or inhibit growth, as intended, and/or provide the appropriate biochemical response. The manufacturer's control checks of media may be used if the product insert specifies they meet the NCCLS standards for media control testing. These individual procedures do not require control checks with each run of patient specimens or further testing unless specified by the manufacturer or under specialty or subspecialty control requirements. Biochemical tests or media that provide microbial identification from a direct specimen or culture (for example, direct antigen tests for group A streptococcus, bacterial serotyping from culture) are considered qualitative microbiology tests and are

graded for reactivity. We are retaining the control procedures requirements for qualitative test systems formerly at § 493.1218(b)(1) (now at § 493.1256(d)(3)(ii)).

*Comment:* One commenter recommended we add "XV discs or strips" to § 493.1227(a)(2) that requires testing both positive and negative control organisms each week of use, and delete § 493.1227(b) that requires testing the XV discs or strips with only a positive control organism each week of use.

*Response:* Testing of XV discs or strips was limited to only a positive control each week of use because there is no known available control to check negative reactivity for the group of organisms that this test identifies. We are deleting the specific QC requirements for testing X, V, and XV discs or strips. These discs or strips are now subject to the general control procedure requirements formerly at § 493.1218(f)(1) (now at § 493.1256(e)(1)) that include testing each new batch (prepared in-house), lot number (commercially prepared), and shipment when prepared or opened for positive and negative reactivity. Since there is no control available to check negative reactivity for XV discs or strips, the use of only a positive control for XV discs or strips will be deemed to meet the CLIA regulation as specified in Appendix C of the State Operations Manual (CMS Pub. 7).

*Comment:* Several commenters recommended we change the control requirement for daily testing of antimicrobial susceptibility procedures to a weekly requirement, as specified by NCCLS. One commenter also suggested manufacturers develop control procedures consistent with NCCLS antimicrobial susceptibility testing standards whenever feasible.

*Response:* CLIA requires daily control checks for antimicrobial susceptibility testing, formerly at § 493.1227(c)(2) (now at § 493.1261(b)(1)) unless CMS approves a procedure that provides equivalent quality testing as specified in Appendix C of the State Operations Manual (CMS Pub. 7). In this case, the procedure providing equivalent quality testing is the NCCLS standard allowing the laboratory to perform weekly control testing of antimicrobial susceptibility procedures after establishing accuracy control limits through initial daily testing. The laboratory may continue performing weekly control testing provided the control results do not exceed the established limits.

*Comment:* One commenter requested clarification of the control requirements for antimicrobial susceptibility testing