Chapter 11
Quality Control of Media and Reagents

Each laboratory must ensure adequate control of the media and reagents it uses. Quality control includes the selection of satisfactory raw materials, the preparation of media according to approved formulations or specific manufacturer's instructions, and the use of well-characterized reference strains to check prepared media.

A. Quality Control of Media

1. Considerations for quality control of media

Each batch of medium prepared from individual ingredients or each different manufacturer's lot number of dehydrated medium should be tested for one or more of the following characteristics, as appropriate:

- Sterility
- Ability to support growth of the target pathogen(s)
- Ability to produce appropriate biochemical reactions

Sterility

One tube or plate from each autoclaved or filter-sterilized batch of medium should be incubated overnight at 35° to 37°C and examined for contaminants.

Ability to support growth of the target organism(s)

- For selective media: use at least one strain to test for ability to support growth of the target pathogen (e.g., for MacConkey agar (MAC), a *Shigella* strain such as *S. flexneri*); it should also be noted if this strain produces the appropriate biochemical reactions/color on the test medium (see below).

Ability to produce appropriate biochemical reactions

- For selective media: use at least one pathogen and one nonpathogen to test for the medium's ability to differentiate target organisms from competitors (e.g., for MAC, a lactose-nonfermenting organism such as *S. flexneri* and a lactose-fermenting organism such as *E. coli*).
- For biochemical media: use at least one organism that will produce a positive reaction and at least one organism that will produce a negative reaction (e.g., for urea medium, a urease-positive organism such as *Proteus* and a urease-negative organism such as *E. coli*).

2. Methods for quality control of media

When testing for ability to support growth, to avoid using too heavy an inoculum, prepare a dilute suspension to inoculate the medium. A small inoculum will

give greater assurance that the medium is adequate for recovery of a small number of organisms from a clinical specimen. An example of a protocol for quality control of media is given here:

- The control strain is inoculated to nonselective broth (e.g., tryptone soy broth) and grown up overnight.
- To prepare a standardized inoculum for testing selective and inhibitory media, make a 1:10 dilution of the overnight nonselective broth culture. If testing nonselective media, prepare an additional 1:10 dilution (to give a 1:100 dilution of the broth).
- One tube or plate of each medium should be inoculated with the standardized inoculum of the control strain(s). When testing selective plating media, a nonselective plating medium such as heart infusion agar should be inoculated at the same time for comparison purposes.
- Using a loopful of the 1:10 or 1:100 dilution prepared above (use a calibrated loop, if available) inoculate media to be tested, streaking for isolation on plating media. The same loop should be used for all quality control of all media; it is more important to use the same inoculating loop every time than it is to use a calibrated loop.

3. Sources of quality control strains

Suitable quality control strains may be obtained in several different ways. A laboratory may use strains isolated from clinical specimens or quality assurance specimens, provided the strains have been well characterized by all available methods (e.g., biochemical, morphologic, serologic, molecular). Many laboratories purchase quality control strains from official culture collections, such as the National Collection of Type Cultures (Public Health Laboratory Service, London NW9, England) or the American Type Culture Collection (12301 Parklawn Drive, Rockville, MD 20852). Quality control strains also may be purchased from commercial companies such as Lab M (Topley House, 52 Wash Lane, Bury, BL9 6AU, England).

B. Quality Control of Reagents

As with all other products used in testing, reagents, either purchased or prepared in the laboratory, should be clearly marked to indicate the date on which they were first opened and the expiration date, if appropriate. Each reagent should be tested to make sure the expected reactions are obtained. If the reagent is a rare, expensive, or difficult-to-obtain product such as diagnostic antiserum, it does not necessarily have to be discarded on the expiration date. If satisfactory sensitivity and specificity can still be verified by normal quality control procedures, the laboratory may indicate on the vial label the date of verification of quality of the reagent. All reagents should be tested for quality at intervals established by each laboratory to ensure that no deterioration has occurred.

Slide agglutination method for quality control of antisera

For quality control of antiserum, two or more control strains (one positive and one negative) should be used to test its agglutination characteristics. The results of all reactions should be recorded. Following is an example of a typical quality control procedure.

- Place a drop (about 0.05 ml) of each antiserum on a slide or plate. Also, place a drop of 0.85% saline on each slide or plate to test each antigen for roughness or autoagglutination.
- Prepare a densely turbid suspension (McFarland 2 or 3; see Table 11-1) of each control isolate in normal saline with growth aseptically harvested from an 18- to 24-hour culture from nonselective agar (for example, heart infusion agar or tryptone soy agar).
- Add one drop of the antigen suspension to the antiserum and the saline. Mix thoroughly with an applicator stick, glass rod, or inoculating loop. Rock the slide back and forth for 1 minute.
- Read the agglutination reaction over a light box or an indirect light source with a dark background. The saline control must be negative for agglutination for the test to be valid.

The degree of agglutination should be read and recorded as follows:

Percent agglutination	Record reaction as:
100	4+
75	3+
50	2+
25	1+
0	negative

C. Advantages of Centralized Acquisition of Media and Reagents

There are several benefits of centralizing acquisition of media and reagents in the national reference laboratory or Ministry of Health:

- Large amounts of a single lot of medium or reagent can be purchased and subsequently divided into smaller aliquots for distribution to provincial/district laboratories. This may be more cost effective (i.e., discount for larger orders, lower shipping costs, less waste because of product going past expiration date).
- Quality control can be performed in the central laboratory, avoiding duplication of effort among provincial and district laboratories. An unsatisfactory medium or reagent may then be returned to the manufacturer before the lot is distributed to other laboratories.
- The standardization of methods among laboratories at all levels is facilitated by use of single lots of media.

Table 11-1. Composition of McFarland turbidity standards

Turbidity standard no.	Barium chloride dihydrate (1.175 %)	Sulfuric acid (1%)	Corresponding approximate density of bacteria/ml
0.5	0.5 ml	99.5 ml	1×10^8
1	0.1 ml	9.9 ml	3×10^8
2	0.2 ml	9.8 ml	6×10^8
3	0.3 ml	9.7 ml	9×10^8
4	0.4 ml	9.6 ml	12×10^8
5	0.5 ml	9.5 ml	15×10^8
6	0.6 ml	9.4 ml	18×10^8
7	0.7 ml	9.3 ml	21×10^8
8	0.8 ml	9.2 ml	24×10^8
9	0.9 ml	9.1 ml	27×10^8
10	1.0 ml	9.0 ml	30×10^8