Judging specimen adequacy by Bethesda ‘01 criteria

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Assessing and reporting specimen adequacy of Pap smears was introduced in the original 1988 Bethesda System. The 1988 Bethesda criteria stipulated that:
- An adequate Pap should contain two groups of five endocervical cells (EC) and/or squamous metaplastic cells.
- At least 10 percent of the slide area should be covered with cellular material.
- Specimens obscured 50 to 75 percent by blood or inflammation should be considered less than optimal, and those obscured greater than 75 percent should be designated unsatisfactory.

In the 1991 Bethesda System revision, the phrase “satisfactory but limited by” replaced “less than optimal.” There was no significant difference, however, in how adequacy was assessed.

Bethesda 2001’s revised adequacy criteria take both published data and theoretical issues into account. The phrase “satisfactory but limited by” has been deleted. The adequacy committee felt the phrase was an oxymoron and that laboratory reports should indicate specimen adequacy less ambiguously. Therefore, Bethesda 2001 designates specimen adequacy as “satisfactory” or “unsatisfactory.”

Specimen quality indicators such as the presence or absence of a transformation zone component, or of obscuring inflammation or blood, are reported after the adequacy designation.

Bethesda 2001’s most significant change lies in the assessment of squamous cellularity. The previous 10 percent coverage criterion can be interpreted in at least two ways: that cellular material covers at least 10 percent of the area under the coverslip (125 mm²) at a “normal” or “usual” cell density, or that 10 percent or 125 mm² of the slide is actually covered or obscured because of the presence of cells. The latter criterion actually represents very high cellularity, and many, if not most, Pap would fail to meet this criterion. Because of the 10 percent criterion’s ambiguity, more specific criteria were developed. For conventional Pap smears, an adequate smear contains between 8,000 and 12,000 well-preserved, well-visualized cells. Labs should not attempt to count cells manually. Several images depicting low-power (~4×) microscope fields with a known number of cells will be available in the updated Bethesda System Manual and on a Web site, so that cytologists can see what a field with, for example, 500 cells actually looks like (Figs. 1 and 2). From these images, estimate overall cellularity.

By Bethesda 2001 criteria, adequate liquid-based prep (LBPs) contain at least 5,000 well-visualized squamous cells. Optionally, labs can elect to append a QI statement indicating borderline cellularity for cases that have 5,000 to 20,000 cells. LBP cellularity is set at a lower level because the number of cells in an LBP can be estimated more easily than the number in a conventional smear, since the cells in most LBPs are distributed more evenly than they are in a conventional prep. Estimate the number of cells in an LBP by counting the number of cells in each of 10 40× fields taken across a diameter of the preparation, and then averaging the results. Some LBPs will have sparsely cellular areas, or “holes,” within the circle. If such holes appear in the preparation, and then averaging the results. Some LBPs will have sparsely cellular areas, or “holes,” within the circle. If such holes appear in the preparation, the 10 40× fields should include a proportionate number of fields from the sparsest cellular areas. Similarly, if portions of the preparation are obscured by inflammation or blood, a proportionate number of the 10 fields should be from those areas. Table 1 provides the number of cells you can expect to find in a 40× field of an LBP with 5,000 total cells for common microscope parameters and prep diameters. With the information in the table, estimating LBP cellularity is not difficult or time-consuming.

For example, with CytoThin Preps, which have a 20-mm diameter circle, and a microscope with field number 22 eyepieces with the 40× objective, approximately four cells per field are necessary to have 5,000 cells on the whole slide. The 40× objective is recommended because the number of cells to be counted is low. Most LBPs contain many more than four cells per 40× field. Therefore, you can determine at a glance whether cells must be counted to check for acceptable cellularity, and only LBPs appearing to have borderline cellularity must be counted.

Before the Bethesda 2001 conference, interested parties used an Internet bulletin board to comment on issues pertinent to the project. Some questioned the necessity of a more rigorous standard for squamous cellularity. Admittedly, relatively little data on which to base a specific quantitative guideline are available, but the committee felt bringing consistency to cellularity assessment through a quantitative standard was important.

Adequacy criteria for obscuring factors have not changed. The criteria for cells from the transformation zone have been modified slightly to require 10 EC or metaplastic cells instead of two groups of five cells each. More studies on EC have been...