WBC LEUKOCYTE DIFFERENTIAL DATA PROCESSING:
An analysis of the merits of approaching assessment of leukocyte differential values by regarding only quantitative absolute counts

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BACKGROUND: Historically, laboratories reported percentage values for the various leukocyte subsets because this is what was actually measured. Manual microscopic counts based on enumerating the fractional components of each type of leukocyte in 100 or 200 cells on a properly stained blood film was the mainstay for many, many decades. The routine method for measuring the WBC count was also manual and much more labor intense with lower analytical precision than current automated methods. As a consequence, percentage values were preferred and WBC counts were relied on less for assessing temporal changes than simply observing proportional shifts in the leukocyte differential results. Much clinical experience was assimilated based on this approach to interpreting proportional leukocyte differential results and on which many clinical correlates and uses were established including recognition of the granulocytic predominance in bacterial infections, lymphocyte predominance in viral infection, significance of high proportion of eosinophils, as well as the significance of a granulocytic “left-shift.” Derivation of quantitative absolute counts was less often considered in the early days for reasons including that WBC counts were not always routinely performed with every differential and both manual differential and WBC methods had relatively high levels of imprecision that would further compromise confidence in the mathematical product of the two values.

It was not until after the introduction of automated WBC and the Coulter particle counter in the 1960s that interest and importance of quantitative differential results rose. Allergists were some of the first physicians to recognize the limitations of percentage values, particularly for cell type that normally comprise a small fraction of the total. In the past they insisted upon quantitative absolute eosinophil counts not derived from the product of the WBC and percentage of eosinophils but from extended counting of stained eosinophils in hemocytometer chambers as a means of improving precision. The ANC became the mainstay of hematologist, oncologists and physicians concerned with marrow recovery and risk of infection. Immunologists and infectious disease specialists concerned about underlying immunodeficiency and infection risk paid special attention to absolute lymphocyte counts as important indicators.

Subsequent to decades of technical improvement, automated WBC counts are exceptionally precise, relatively inexpensive, fast to perform and seldom need confirmed by alternative methods. The 1980s saw the introduction of an automated “3 part” leukocyte differential followed by a 5 part and now up to 6 part automated differentials that offer superior levels of precision by routinely counting thousands of cells and do not have the high levels of inter-observer bias inherent in manual microscopic based differentials. The microscope based manual assessment still finds limited use in special circumstances including screening smears for qualitative anomalies, generating differential results when the instruments cannot, detection of rare cell types and used in instances where extended differential data on infrequent cell types is needed. However, most leukocyte differentials today are automated in most laboratories including pediatric facilities. Because clinical hematology instrument differentials are based on counting fixed volumes of sample, quantitative absolute differential cell counts are in fact the “measured” parameter and differential percentage values are now calculated from the absolute counts. Surprisingly in spite of these advances and greater simplicity of reviewing numbers that are quantitative, physicians today still prefer seeing leukocyte differential percentage data over absolute cell counts, probably as a result of being taught in the fashion 1. Dealing with percentage data is still being taught to medical students and house staff and has a firm foothold in the United States and other nations though the trend is not universal.

APPROACHES TO PROCESSING DIFFERENTIAL DATA: The common approach employed by most generalist and primary care specialists using percentage values places primacy on the WBC count and identifying the cell fraction that accounts for directional shift in cases were the WBC count is abnormal. For example: In a patient with abnormally high WBC count, interest is directed to detecting the cell fraction with elevated percentage (e.g. neutrophils of 82% which would lead to the conclusion that the patient has a granulocytosis supporting a clinical suspicion of bacterial infection). Alternatively, if the WBC is normal, then the percentage values for each of the component cell types is compared to reference range percentage values to identify an abnormality.
The advantage of this approach is said to be that normal percentage ranges for all the component cell types are easier to remember than quantitative ranges. The disadvantage is that minor cell alterations that are not large enough to swing the WBC may carry clinical significance and are easily missed. Furthermore, reference normal percentage ranges are not applicable when the WBC is abnormal and are not necessarily applicable even when the WBC is normal. On certain occasions when the WBC is in the normal range, percentage values fall within their stated “normal” ranges when in fact they are quantitatively abnormal. The latter may occur when percentage for lymphocytes are near the low end of “normal” while the neutrophils are on the high end of “normal” increasing the chances of missing either a granulocytosis when the WBC is toward the high end of the normal range, or missing lymphopenia when the WBC is at the low end of the normal range. In the past when WBC and differential values were inherently imprecise, minor deviations were often ignored because it was not possible to attribute them to disease as opposed to reflecting method imprecision. This is not the case when dealing with today’s testing methods described above; minor deviations are important indicators of underlying disease. It has been shown that even neutrophil values within the upper end of the normal range are indicators of increased risks of cardiovascular disease compared with counts in the lower range of normal. To reliably detect all aberration, whether WBC is elevated or not, one would need to view absolute counts. Furthermore, various component cell types respond to physiologic and pathologic signals in an independent manner resulting in complex and often independent quantitative deviations from normal in ways that are not proportional.

The differential interpretive approach using only absolute counts places primacy on detecting aberrations in all leukocyte subtypes irrespective of WBC value. The method is direct and as simple as reviewing any other quantitative lab value without the need to bother looking at and processing percentage values. The method simply relies on reviewing only quantitative counts for each cell type and comparing values with established normal reference ranges. This approach is not dependent on a complex multi-step data processing algorithm described using percentage values or prone to processing errors. Unlike percentage values, absolute normal range values are independent of WBC count and recognizing deviations from normal in all cell line is straightforward. Furthermore, quantitative numbers are more reliable for tracking temporal changes in the course of disease or in response to treatment. Tracking temporal changes in percentage value for a given cell type can be misleading when the fractional percentage rises or falls disproportionate to (and sometimes in opposite direction from) the corresponding absolute count. For example: in an infected patient the percentage of neutrophils may temporarily rise at the same time as the absolute neutrophil count falls as a result of the sudden lympholytic effect of added steroid therapy, potentially leading one to falsely conclude on the basis of percentage values alone that there may be a worsening infection or raise inappropriate concern about ineffective antibiotic therapy. This type of error would not arise following absolute counts where recognizing both the sudden lymphopenia and falling neutrophil count is straightforward, reliable and not subject to errors inherent with processing percentage values. Disadvantages of this method is that it is hard to remember normal absolute ranges for all the various component cell lines and user difficulty conceptualizing numerical values that may involve 2 or more decimal points (e.g. immature granulocytes = 0.090 x 10^6 / microL).

Regarding only the major cell type responsible for shift in the WBC seldom has consequences because the other cell types are either normal or seldom is there a clinical consequence when they are abnormal. However, significant underlying diseases may be overlooked with adverse economic and patient outcome. Delays in diagnosis of SCIDS with death, extended hospitalizations and worse outcome post transplantation results not uncommonly from overlooking lymphopenia in patients who commonly presents with leukocytosis due to granulocytosis associated with an opportunistic infection. Even if there is not an underlying immunodeficiency, lymphopenia in sick infants is important and has been shown to be associated with worse disease and probably should not be ignored. The common use and approach to percentage data also leads to missing significant aberrations in cell numbers when the overall WBC is normal as described above, perhaps a reason to consider dropping WBC counts from reports when absolute leukocyte differential data is presented. It is also clear that many busy clinicians have resorted to looking only for laboratory high or low flagged values, an effort that would be more productively spent on quantitative values. Other errors related to regarding percentage values include delayed diagnosis of Chronic Lymphocytic Leukemia and over or underestimating eosinophilia. Sometimes these errors leads to inappropriate investigations with added costs or are used inappropriately to support a diagnosis of atopic or other illnesses. These types of errors are unlikely to occur when dealing with only absolute counts. Other examples exist along these lines.
The transition to regarding only the absolute count takes some effort and perhaps a leap of faith by older physician. Many developed countries of the world including Canada, Scandinavia and Great Britain do not report percentage values (personal communications). Dropping the percentage values would bring focus to the important quantitative differential parameter and help avoid sending newly trained resident off to continue using outdated interpretive approaches more prone to error and which add unnecessary length to the medical records.

References:

2. Armin J. Grau, MD; Alexander W. Boddy, MS; Deborah A. Dukovic, MS; Florian Buggle, MD; Christoph Lichy, MD; Tobias Brandt, MD; Werner Hacke, MD; for the CAPRIE Investigators. Leukocyte count as an independent predictor of recurrent ischemic events. Stroke 2004;35:1147-1152
4. Myers LA, Patel DD, Puck JM, Buckley RH. Hematopoietic stem cell transplantation for severe combined immunodeficiency in the neonatal period leads to superior thymic output and improved survival. BLOOD. 2002;99:872-878.

July CME has been canceled.