LETTER TO THE EDITOR

ESTIMATION OF SAMPLING ERRORS

July 26, 1984

The article by H. Harms and H.M. Aus (1) contains a mistake in the theoretical development of their work. While the mistake is fundamental, the overall results, developed in the article and summarized in Figure 4, will be altered only slightly.

The mistake is to be found in equation 1 and then repeated in equations 2 and 7 and Figure 3. The mistake is a misinterpretation of the system diagram in Figure 1. By itself, the diagram is correct; every subsystem that is linear, shift-invariant can be represented either by its point-spread function (impulse response) or its Fourier transform. What is important to realize in the inner-connection of the subsystems is whether the relationship between the output variables of one box and the input variables of the next box should be specified in the space domain or the frequency domain. For example, the relationship of the first three subsystems is clearly given by $F(w) = I(w) \times O(w) \times L(w)$, that is, the multiplication of the three Fourier spectra representing the convolution of their respective space-varying functions $i(x,y)$, $o(x,y)$, and $l(x,y)$. The diagram relating the variables is in agreement with standard practice for such system diagrams.

The first of two errors, associated with misinterpretation of Figure 1, develops with the authors' representation of the finite aperture effects. Generally, aperture effects occur in the spatial (or time) domain. That is, only a portion of the input signal can be analyzed and the rest must be thrown away. If this, indeed, is the model the authors suggest, then the correct mathematical relationship relating the signal $f(x,y)$ and the aperture function $b(x,y)$ is given by the following:

$$f(x,y) \ast b(x,y) \quad (1a)$$

(multiplication in the spatial domain with truncation by windowing function $b$)

or expressed in the Fourier domain as

$$\left(\frac{1}{2\pi}\right) F(w) \ast B(w) \quad (1b)$$

(convolution in the frequency domain).

The convolution given above is in agreement with their equation 1 and their subsequent neglect of the aperture effect that results in equation 2. This formulation, however, is not correctly depicted (according to standard notation) in Figure 1. What is depicted in the system diagram in Figure 1 is a convolution of $f(x,y)$ and $b(x,y)$, at least according to standard notation. Thus, according to their system diagram, the relationship between $f$ and $b$ is given by

$$f(x,y) \ast b(x,y) \quad (2a)$$

(convolution in the space domain)

which is equivalent to

$$F(w) \ast B(w) \quad (2b)$$

(multiplication in the frequency domain with truncation by windowing with $B$).

This first error now is clear. While equations 1a and 1b above are in agreement with their equation 1, equations 2a and 2b above are in agreement with their Figure 1. The two sets of equations definitely are not the same! Further, it is possible that the system diagram (and thus equations 2a and 2b above) represent the desired model for the aperture effect. If the authors are referring to a possible aperturing in the back focal-plane of the microscope objective lens (under near monochromatic illumination conditions), then the second set of equations represents the truncation of the Fraunhofer (Fourier) diffraction pattern and thus an equivalent convolution in spatial domain terms. In their article, the authors never make it clear where in the optical chain this aperturing occurs and thus it is not possible to decide which model is the appropriate one. In any case, there is a clear inconsistency between their equation 1 and their Figure 1.

This particular error plays no further role in their paper, as the authors choose (quite logically) to ignore the effects of aperturing in the development of their theory. The fundamental mistake of misinterpreting the system diagram does play a role, however, in a second and significantly more important mistake.

The authors (correctly) show the sampling process in Figure 1 as the multiplication of the image by a periodic pulse train with a specific (in this case Gaussian) shape. Thus, if the image (neglecting the aperture effect) is given by $f(x,y)$ and the sampling function by $s(x,y)$, then the sampled image (in their terms) is given by

$$h(x,y) = f(x,y) \ast s(x,y) \quad (3a)$$

(multiplication in the spatial domain)

which is equivalent to

$$H(w) = \left(\frac{1}{2\pi}\right) F(w) \ast S(w) \quad (3b)$$

(convolution in the frequency domain).
In equations 1, 2 and 7, the authors show an incorrect expansion of regrouping of the terms appearing in equation 3b above. The correct modeling of the effect of the *sampling aperture* \(a(x,y)\) is given by

\[
s(x,y) = a(x,y) * p(x,y) \quad \text{(space domain)}
\]

\[
S(w) = A(w) X(w) \quad \text{(Fourier domain)}
\]

where \(A(w)\) and \(X(w)\) are the signals defined immediately after equation 1 in the article. Thus, the sampled signal in the frequency domain can be rewritten as

\[
H(w) = \left( \frac{1}{2\pi} \right) F(w) * [A(w) X(w)].
\]

This is not the same as their equation 2. Further, because \(X(w)\) represents a train of ideal impulses, the term \([A(w) X(w)]\) can be written as

\[
\sum_{k=-\infty}^{+\infty} A(kw_o) F(w - kw_o).
\]

Finally, equation 5 (above) can be written as

\[
H(w) = \left( \frac{1}{2\pi} \right) \sum_{k=-\infty}^{+\infty} A(kw_o) F(w - kw_o).
\]

In words, the following occurs: The act of sampling with a nonideal aperture means that an infinite number of copies of the original image spectrum \(F(w)\) are generated (periodically spaced) but each with a different amplitude. There is no change in the form of the spectrum in each of the periodic copies, only the amplitude (scale) factor. Thus lines c and e in Figure 3 are wrong. The baseband \((k = 0)\) term in the spectrum of \(H(w)\) that is, the term between the normalized frequencies \(-1\) and \(+1\), should be

\[
H(w) = \left( \frac{1}{2\pi} \right) A(0) F(w) = \left( \frac{1}{2\pi} \right) (1 - w/w_{max}),
\]

where \(A(0) = 1\) according to the assumption used by the authors immediately following equation 7 in their article (1). Note the significant difference between equation 8 above and the incorrect version given as equation 7, in the article (1), which describes the effect of the *scanning*, not sampling, aperture.

The effect that this correction will have upon recalculation of Figure 4 is difficult to judge. My estimate is that the difference will be small; that is, Figure 4 and the conclusions drawn from it, will not change much. The dropping of the Gaussian term in their equation 7 probably will mean one rescaling of the data for the 10% cutoff values and a second rescaling of the data for the 50% cutoff values.

A more detailed explanation of the theoretical background behind this aspect of sampling theory is given in Oppenheim, Willsky, and Young (2,3).

As a trivial additional point, I believe that there is a "typo" in equation 9 (1) and that "ld" should be "ln", that is, the logarithm function.

I hope my comments will be useful to the authors and to the readers of Cytometry.

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**LITERATURE CITED**


**REPLY TO DR. YOUNG**

October 23, 1984

The main points in Dr. Young’s letter are 1) a lack of standard notation in the system block diagram, 2) a correction in the equations and analysis which describe a sampling system, and 3) a typo in the information content equation 9.

We appreciate Dr. Young’s comments and the opportunity to clarify these points, which we address specifically.

1) System block diagram modification. The intent of Figure 1 in our paper (1) is to show which optical components and sampling methods contribute to the system’s response and characteristics. A modified system block diagram, using the standard notation suggested by Dr. Young, is shown here as Figure 1 (modified). The major difference is the explicit inclusion of the two convolution operators.

![System Diagram](image)

**FIG. 1 (modified). System diagram (see reference 1).**