The following full text is a publisher's version.

For additional information about this publication click this link.
http://hdl.handle.net/2066/22757

Please be advised that this information was generated on 2020-02-13 and may be subject to change.
PRECISION AND ACCURACY OF ACOUSTOSPECTROGRAPHIC PARAMETERS

H. J. HUISMAN and J. M. THIJSSEN

Biophysics Laboratory, Department of Ophthalmology and Clinical Physics Laboratory, Department of Pediatrics, University Hospital, Nijmegen, The Netherlands

Abstract—Theoretical estimates of the standard deviation (STD) of four acoustospectrographic parameters (the intercept and slope of attenuation and backscatter coefficient) are derived. This derivation expands and corrects existing derivations, and is confirmed using simulations based on the adopted theoretical model. A robust parameter estimation method is applied to various phantom measurements, and to in vivo liver scans of healthy human subjects. The measured STD is higher than the theoretically predicted value, and we investigated four possible factors which explain this discrepancy. First, it is shown that the STD and bias after spectrogram calculation are rather insensitive to changes in windowing function, type, length and overlap. Second, we observed that a diffraction correction spectrogram calibrated on a medium different from the one being measured insufficiently corrects the depth-dependency of the parameters, which affects both precision as well as accuracy. We therefore propose a method that constructs an organ-specific diffraction correction spectrogram from the averaged spectrogram of a set of normal organs. We show that the organ-specific correction does not affect STD even in case of previously unseen acquisitions. Third, we introduce local inhomogeneity, which predicts excess STD due to local variations of the physical parameters within an organ (i.e., intrasubject), and global inhomogeneity, which predicts variations between organs (i.e., intersubject). We conclude that our method of estimating STD predicts normal, in vivo data very well, and propose that the deviation from these estimates is a potential tissue characterization parameter.

Key Words: Tissue characterization, Attenuation, Backscatter, Precision, Accuracy, Diffraction correction, Inter- and intrasubject variability.

INTRODUCTION

The radio-frequency (RF) echo spectrum has been used extensively in medical ultrasound research to interrogate noninvasively the structural properties of biological media. One approach is to use the frequency dependence of the RF backscatter spectrum. Many strategies exist for attenuation and backscatter measurements (Cloostermans and Thijssen 1983; Insana et al. 1983; Kuc et al. 1976; Kuc and Schwartz 1979; Lizzi et al. 1976; Lizzi et al. 1983; Mountford and Wells 1972; Nicholas et al. 1982; Ueda and Ozawa 1985). In this article, attention is focused on the precision (described by the standard deviation [STD]) and the accuracy (described by the bias) of four acoustospectrographic parameters: the intercept at the central frequency and the slopes of the attenuation and backscatter coefficients.

A detailed understanding of the precision and accuracy of the parameters is important for three reasons. First, the required precision gives a theoretical minimum of the scanned volume required for differentiation between normal and malignant tissue. Second, in theory, bias is zero and STD can be estimated very well, but, in practice, various factors affect accuracy as well as precision. Insight is gained into the relative importance of each of these factors, which can be used for focusing research on improvement of methods. Finally, one of the factors is shown to be a possible new tissue characterization parameter.

Various sources of error complicate the parameter estimation process. A trivial source is the effect of the overall gain and the time gain control (TGC) settings on the echographic equipment when using the output video.
signal. We ruled out these dependencies by acquiring the internal, unprocessed RF signal from within the equipment (custom-built RF interface) using a custom-designed RF acquisition workstation (Verhoeven and Thijsen 1992). The most important source of error is the speckle noise inherent to the backscattering process found in many biologic media. The effect of the speckle noise can be estimated theoretically. The following sources of error cannot be estimated theoretically, but it is possible to obtain practical estimates. The depth-dependent sound field and the electroacoustic pulse spectrum introduce errors in the estimation process which increase the STD of the estimates. We will show a method that enables adequate correction and we will quantify the increase in STD due to the correction. The acoustospectrographic parameters measure local medium parameters assuming homogeneity conditions. In practice this appears not to be realistic, and thus a local inhomogeneity factor is introduced which measures the variation due to variation in local medium parameters. When measuring liver tissue, a global inhomogeneity, in addition to the local inhomogeneity, is also observed, which is due to the individual spread of parameter values.

An expression for the STD of the attenuation slope (one of the four parameters studied in this article) has been the topic in a number of articles (Berger et al. 1987; Kuc 1985; Kuc and Taylor 1982; Parker 1986; Romijn et al. 1989). Yao (1991) also analyzed the STD of the backscatter parameters. The formulas only describe STDs under theoretical conditions and vary due to different levels of approximations and parameter estimation methods. We have derived formulas for the STD of all four parameters, using as few approximations as possible. We show, with simulated RF data, that they accurately predict the STD of each parameter over a wide range of measurement conditions. Finally, we analyze a set of in vivo normal liver measurements. We arrive at an estimate of the local and global inhomogeneity in the liver and hypothesize on using the local inhomogeneity as a new parameter.

This article is organized as follows. First, the theoretical signal model is described which is subsequently used in the second section to present the parameter estimation method. The third section uses both the model and the method to derive the STD of the parameters. In the fourth section, attention is focused on the transducer-dependent deviations: measurement of transducer characteristics (TC) is described, as well as the effect of correction on the parameter deviations. The Results section presents STDs observed in phantom and normal in vivo liver data. Finally, conclusions are summarized and future work is discussed.

**Theory**

Assuming a homogeneous tissue model, the amplitude spectrum of the backscattered echo signal received in pulse-echo mode, \( E(f, z) \), is a random variable (indicated in bold) with Rayleigh-distributed amplitudes, and an average value, \( \mu(f, z) \), that can be described by a linear model (Oosterveld et al. 1991):

\[
Pr \{ E(f, z) = E(f, z) \} = \frac{\pi E(f, z)}{2\mu^2(f, z)} \exp \left( -\frac{\pi E^2(f, z)}{4\mu^2(f, z)} \right) \quad (1)
\]

\[
\mu(f, z) = P_2(f) D^2(f, z) T^2(f, z) S(f) \quad (2)
\]

where \( E(f, z) \) is the received amplitude at temporal frequency \( f \) of the backscattered echoes of a small tissue volume at depth \( z \), \( P_2(f) \) is the acoustoelectric transfer function of the transducer, \( D^2(f, z) \) is the diffraction spectrogram of the transducer, \( T(f, z) \) is the tissue transfer function, and \( S(f) \) is the backscatter function.

The terms \( P_2(f) \) as well as \( D^2(f, z) \) in eqn (2) jointly describe the transducer characteristics (TC). Without a proper correction procedure, they will affect the parameter estimates (Cloostermans and Thijsen 1983). The Transducer Characteristics Estimation section describes TC measurement methods capable of estimating both terms. After correction, the remaining two terms in eqn (2) are \( T^2(f, z) \), and \( S(f) \), which describe the tissue. They are developed further to show how the four tissue parameters are involved in the model.

A general expression for the tissue transfer function is:

\[
T(f, z) = 10^{-a(f)/2 \times 10} \quad (3)
\]

where \( a(f) \) is the attenuation coefficient [dB/cm]. Pauly and Schwan (1971) showed that the attenuation coefficient could be approximated by a linear model. The coefficients of the linear model in a limited frequency range can be shown to produce an estimate of a causal relaxation model (Berkhoff et al. 1996; Jongen et al. 1986). We use the following, linear model:

\[
a(f) = a_0 + a_1(f - f_c) \quad (4)
\]

where \( f_c \) is the central frequency in the available bandwidth, which are both fixed parameters dependent on the echographic system, \( a_0 \) is the attenuation intercept.
at the central frequency [dB/cm], and $a_i$ is the attenuation slope [dB/cm MHz].

A general expression for the backscatter function is:

$$S(f) = 10^{0.05b(f)}$$

(5)

where $b(f)$ is the backscatter coefficient [dB]. In a limited frequency range, the backscatter coefficient exhibits quasi-linear shapes (Lizzi et al. 1987; Romijn et al. 1989), and can thus be approximately described by:

$$b(f) = b_0 + b_1(f - f_i)$$

(6)

where $b_0$ is the backscatter intercept at the central frequency [dB], and $b_1$ is the backscatter slope [dB/MHz].

The Rayleigh probability density function (pdf) is due to the assumed microstructure of the medium: many small (compared to the wavelength) and randomly positioned scatterers. The received backscattered echo signal at the surface of a piezoelectric transducer is the result of the phase-sensitive addition of randomly phased backscattered echoes from many, small scatterers from this microstructure. The amplitude of the received signal is the result of the classical random walk problem, which was shown to produce Rayleigh-distributed amplitudes in the cases of laser light (Goodman 1975) and ultrasound (Burckhardt 1978; Wagner et al. 1983).

**Medium-dependent transducer characteristics**

The model, as presented in eqn (2), assumes that the diffraction spectrogram, $D(f, z)$, is separable from the medium backscatter characteristics, $S(f)$. If this is true, then the diffraction spectrogram measured in an artificial medium is equal to that measured in tissue. The latter observation has been used by many researchers in measuring the diffraction spectrogram using plane reflectors (Lizzi et al. 1983; Madsen et al. 1984; O'Donnell, 1983), using foam phantoms (Cloostermans and Thijssen 1983; Fink et al. 1983), or analytically (Insana et al. 1994). However, Robinson et al. (1984) reported that: “corrections obtained from T.M. [tissue-mimicking] phantom material differ from those obtained using in vivo tissue as the reflector.” They compared flat plate, small scatterer phantoms, and in vivo diffraction spectrograms and found that they differed markedly. Laugier et al. (1987) conclude that: “... the best accuracy is reached ... if the transducer is calibrated in vivo on the organ with unknown attenuation.” Céspedes and Ophir (1990) also acknowledge the problem and use a special transducer setup to acquire simultaneously signal as well as estimating diffraction through a so-called axial beam translation technique.

A good example of the dependency of measured diffraction on the medium was shown by Verhoef et al. (1985). They carried out simulations using single scatterer, multiple random scatterers, and a plane reflector. Their results are shown in Fig. 1. The diffraction effect measured from a single scatterer is shown to coincide very well with the average backscatter spectrum of a cloud of random scatterers. The apparent diffraction measured with a plane reflector markedly deviates from the single scatterer curve. They concluded: “it will be clear that the use of flat plate reflections for the estimation of the diffraction factor is incorrect in backscatter estimation.”

It will be shown that the diffraction spectrogram measured on foam phantoms allows for depth-independent measurements on independent foam data. The same diffraction spectrogram when used on in vivo liver data is shown not to provide adequate correction. Using a diffraction spectrogram obtained from in vivo data did result in depth-independent measurements.

### PARAMETER ESTIMATION METHOD

An essential feature of any biomedical measurement system is that it has to tackle the in vivo measurement conditions. That is, clinically realistic measured echo data is affected in many areas by big and small blood vessels, ligaments and acoustic shadowing that are not included in the homogeneous signal model. These areas drastically affect the estimated parameters and should thus be avoided. We incorporated a strategy throughout our parameter estimation method to avoid these areas. We use a mask matrix in which it is indicated for each sample to which area it belongs, and any sample not marked belongs to homogeneous tissue of the organ of interest. The mask matrix is initially filled manually by outlining: the organ boundary (samples outside the organ are marked), and unwanted structures within the organ, such as major blood vessels. An overflow detection step then marks those samples that were out of range in the A/D conversion step. Then, an iterative detection technique automatically marks hyper- or hypoechoic windows (average signal power in the window is outside a 95% reliability interval, which is estimated over all windows in the organ). Underflow areas are excluded by marking windows that have a low signal-to-noise power ratio. Finally, a small blood-vessel detection method extends the detection capabilities to detect small structures which could easily be missed during manual outlining, and are in the order of the window size,
which makes them difficult to detect using a spectral window. The detection of these small structures requires one extra iteration. The attenuation is first estimated with the available mask, and attenuation and diffraction are then used to correct each acquired RF line, which are then envelope-detected. The resulting envelope image is smoothed and then thresholded. This results in the required mask. The smoothing window size determines the size of the detected structures.

The estimation starts by calculating the spectrogram of each RF line in a selected region-of-interest (ROI). An average spectrogram is calculated by averaging over all lines in the ROI, excluding the windows in a spectrogram that contain any marked samples in the mask matrix. The average spectrogram is denoted by $E(f, z)$.

The estimation then continues by removing the estimated transducer characteristics (see Transducer Characteristics Estimation section) from the average spectrogram. After division of $E(f, z)$ by the two terms comprising the estimated $TC$ ($\hat{B}^2(f), \hat{D}^2(f, z)$), an estimated tissue transfer function and backscatter function ($\hat{T}^2(f, z), \hat{S}(f)$) remain.

The next step is to log transform the TC-corrected, average spectrogram. Using eqns (2), (3) and (5), the result can be written as follows:

$$LME(f, z) = 20 \log_{10} \left( \frac{E(f, z)}{\rho^2(f) \hat{D}^2(f, z)} \right)$$

$$= 20 \log_{10}(\hat{T}^2(f, z) \hat{S}(f))$$

$$= 2\hat{a}(f)z + \hat{b}(f) + \epsilon \quad (7)$$

where $LME$ is a random variable resulting from the log of the mean of the spectrogram, the operator $\wedge$ indicates the estimated variable, and $\epsilon$ is a zero mean, Gaussian random variable, which will be shown in the next subsection.

Attenuation is estimated using the method described by Cloostermans et al. (1983) and Fink et al. (1983). A least-squares straight line is fitted with depth for each frequency of the spectrogram $LME(f, z)$. As can be observed from eqn (7), the slope is an estimate of the attenuation coefficient, and the intercept is an estimate of the backscatter coefficient. This method will be referred to as multi-narrow band (MNB). The estimated attenuation coefficient, $\hat{a}(f)$, is used again in a second linear fit, which results in an estimate of both parameters from eqn (4). The resulting backscatter coefficient, $\hat{b}(f)$, is also used in a second linear fit, which results in an estimate of both parameters from eqn (6).

The frequency bandwidth in which the parameter
estimation methods accurately operate is limited by the digitizing system, and a noise floor from the electronic amplifiers (Kuc 1985). We set the TGC amplifier such that the average amplitude of the digitizer output signal at each depth was always between 50% and 100% of the maximum output value. Under these conditions, we observed from the STD estimates of the attenuation coefficient that the estimation methods best operate within a bandwidth such that all frequency components at all depths are within —12 dB of the maximum spectral amplitude of the RF spectrogram before TGC correction.

### STD of Attenuation and Backscatter Parameters

With the signal model and the estimation method being available, it is now possible to derive the STDs and discuss any bias of the parameter estimates. First, the STD of the estimated LME spectrogram is derived. This result is then used to derive the STDs of the attenuation and backscatter coefficient. The STD of the fit to the coefficients then leads the STD of slope and intercept. This section concludes with an investigation of factors influencing STD in practice.

### STD of log average amplitude spectrogram

The amplitude spectrum, $E(f, z)$, is modeled as a random variable with a Rayleigh pdf (see Theory section). For Rayleigh pdf, it can easily be shown that (Papoulis 1991):

$$\sigma_{E(f,z)} = \mu_{E(f,z)} \sqrt{\frac{4 - \pi}{\pi}} \approx \mu_{E(f,z)}/1.91 \tag{8}$$

where $\mu_{E(f,z)}$ and $\sigma_{E(f,z)}$ are, respectively, the mean and the STD of the echo amplitude at frequency $f$. To decrease the STD of the parameter estimates, the average amplitude spectrum, $\bar{E}(f)$, is used. If $N$ independent lines are averaged, then, if $N$ is large enough, the resulting average spectrogram is again a random variable, but with a Gaussian pdf, due to the central limit theorem (Papoulis 1991), and mean and standard deviation:

$$\mu_{\bar{E}(f,z)} = \mu_{E(f,z)} \tag{9}$$

$$\sigma_{\bar{E}(f,z)} = \sigma_{E(f,z)}/\sqrt{N} = \mu_{E(f,z)}/(1.91\sqrt{N}) \tag{10}$$

As was shown in the previous section, the parameters are estimated from the log transform of $E(f)$. If $N$ is large enough, the STD is small compared to the mean, and the STD of the log-transformed variable is estimated using the first order Taylor approximation of the log operator ($20 \log_{10}(x + \Delta) \approx 20 \log_{10}(x) + 8.69\Delta/x$). Thus, the mean and STD of the log transformed variable become:

$$\mu_{LME(f,z)} = 20 \log_{10}(\mu_{E(f,z)}) = 2\sigma(f)z + b(f) \tag{11}$$

$$\sigma_{LME(f,z)} = 8.69\sigma_{E(f,z)}/\mu_{E(f,z)} = 4.54/\sqrt{N} \tag{12}$$

The STD of the log average spectrogram has become independent of the average value, and is a constant that depends on $N$ only, $\sigma_{LME(f,z)} = \sigma_{LME}$. The log average transform thus results in a homoscedastic random variable with additive, Gaussian noise. Equation (12) is equivalent to that by Parker [1986, eqn (20); he used the natural logarithm].

### STD of the attenuation and backscatter coefficient

Assume $LME(f_k, z_i)$ contains $L$ independent spectra ($z_i, 1 \leq i \leq L$), and $K$ discrete, independent frequencies ($f_k, 1 \leq k \leq K$). A linear least-squares fit to $LME(f_k, z_i)$ with depth is optimal, because [see eqn (7)] $\epsilon$ is an additive Gaussian noise term. Under such conditions, it can be shown that the attenuation and backscatter coefficient estimates also have a Gaussian distribution (Kleinbaum et al. 1988). Assuming $L$ large enough, the STD of the attenuation and backscatter coefficient is very well approximated by (see Appendix A):

$$\sigma_{a(f)} \approx \sqrt{\frac{3\sigma_{LME}}{D/L}} \text{ [dB/cm]} \tag{13}$$

$$\sigma_{b(f)} \approx \frac{2\sigma_{LME}}{\sqrt{L}} \sqrt{1 + 3\delta + 3\delta^2 - \frac{3 + 6\delta}{2L}} \text{ [dB]} \tag{14}$$

where $D$ is the ROI length ($D = \Delta z L$), and $\delta$ is the relative delay to the first spectrum ($= z_1/D$), and $\sigma_{LME}$ is given by eqn (12).

The STD of the backscatter coefficient thus decreases with an increasing ROI length, and increases with an increasing delay $\delta$. The latter term within the larger square-root sign is a correction term that is effective whenever the number of spectra is low and/or the relative delay is high. As is seen from eqns (13) and (14), the STDs are independent of frequency, thus $\sigma_{a(f)} = \sigma_a$, and $\sigma_{b(f)} = \sigma_b$.

The approximations in eqns (13) and (14) were verified by a straightforward simulation of the model.
Fig. 2. Simulated (data points) vs. predicted (solid line) STDs of the attenuation and backscatter coefficients. Each data point is estimated from 100 realizations, $\sigma_{\text{LME}} = 1$. As expected, the STD of the attenuation coefficient depends on $L$ as well as $D$, but not on the delay ($M$ is the delay expressed in number of windows). The backscatter coefficient depends on $L$ as well as the delay, but not on $D$.

specified in eqn (7). Figure 2 shows the predicted (solid line) and measured (data points) STDs for the attenuation and backscatter coefficient. The theoretical predictions fit the simulated measurements very well even at a very low number of windows ($L = 3$)!

**STD of the attenuation and backscatter slope and intercept**

The four parameters are estimated by estimating the slope and intercept from the attenuation and backscatter coefficients. The STD of the coefficients was shown not to depend on the frequency, and again an additive Gaussian model can be used with the accompanying least-squares fit. The four acoustospectrographic parameters from the signal model are estimated in an optimal sense; i.e., zero bias, and minimal STD.

The estimated attenuation coefficient can be modeled as:

$$a(f_k) = a_0 + a_1(f_k - \bar{f}) + \epsilon_a \quad (15)$$

where we assume the central frequency to be the midpoint frequency ($\bar{f}$) in the available bandwidth, and $\epsilon_a$ is an additive Gaussian distributed noise term ($p(\epsilon_a) = N(0, \sigma_a)$). If the number of frequencies is much larger than 1, and using a similar derivation as in Appendix A, the STD of the slope is given by eqn (32).

The intercept is at central frequency, and eqn (29) can directly be simplified to

$$\sigma_{a_0} = \frac{\sigma_a}{\sqrt{K}} \approx \frac{\sqrt{3}\sigma_{\text{LME}}}{DNKL} \quad [\text{dB/cm}] \quad (16)$$

$$\sigma_{a_1} \approx \frac{\sqrt{12}\sigma_a}{W\sqrt{K}} \approx \frac{6\sigma_{\text{LME}}}{DW\sqrt{KL}} \quad [\text{dB/cmMHz}] \quad (17)$$

where $W$ is the bandwidth [MHz] ($W = \Delta fK$). The backscatter coefficient can be modeled as:

$$b(f_k) = b_0 + b_1(f_k - \bar{f}) + \epsilon_b \quad (18)$$

where $\epsilon_b$ is an additive Gaussian distributed noise term ($p(\epsilon_b) = N(0, \sigma_b)$). If the number of frequencies is much larger than 1, then:
\[
\sigma_{b_l} = \frac{\sigma_{b_l}}{L} \approx \frac{2 \sigma_{LME}}{KL} \times \sqrt{1 + 3 \delta + 3 \delta^2 - \frac{3 + 6 \delta}{2L}} \text{ [dB]} \quad (19)
\]

The term \( KL \) can be developed even further. \( D \) and \( W \) are related via the window duration \( T \). Using \( \Delta f = 1/T \), and \( \Delta x = cT/2 \) (no overlap) the product becomes \( KL = \sqrt{DW}/\sqrt{c/2} \), where \( c \) is the speed of sound in the medium. The STD of the slopes and intercepts then become:

\[
\sigma_{a_0} \approx \frac{0.5 \sqrt{6 \sigma_{LME}}}{D \sqrt{DW}} \text{ [dB/cm]} \quad (21)
\]

\[
\sigma_{a_1} \approx \frac{3 \sqrt{2 \sigma_{LME}}}{D \sqrt{DW}} \text{ [dB/cmMHz]} \quad (22)
\]

\[
\sigma_{b_0} \approx \frac{\sqrt{2 \sigma_{LME}}}{\sqrt{DW}} \sqrt{1 + 3 \delta + 3 \delta^2 - \frac{3 + 6 \delta}{2L}} \text{ [dB]} \quad (23)
\]

\[
\sigma_{b_1} \approx \frac{2 \sqrt{6 \sigma_{LME}}}{W \sqrt{DW}} \times \sqrt{1 + 3 \delta + 3 \delta^2 - \frac{3 + 6 \delta}{2L}} \text{ [dB/Hz]} \quad (24)
\]

The above equations were verified by simulation. Figure 3 shows the predicted (solid line) and measured (data points) STDs for the slope and intercept of the attenuation and backscatter coefficient. The predictions fit the simulated measurements very well.

**STD in practice**

We will consider five factors that may influence the theoretical estimates of the STD derived previously. We will select the relevant factors and show how they are used to convert a theoretical STD estimate into a realistic estimate.

The first factor concerns the conversion of RF lines into spectrograms. In theory, the RF spectrogram, \( LME(f, z_t) \), is assumed to be comprised of independent components. In practice, the spectrogram is calculated from the RF time-domain signal, \( e(t) \), and we use the short-term Fourier transform [STFT, or periodogram (Welch 1967)] to do so. In short, the STFT divides \( e(t) \) into \( L \) windows. Window 1 is used to calculate the backscattered amplitude spectrum from a tissue volume at depth \( z_t \). The window is selected from \( e(t) \) by applying a window function \( w(t) \) at \( t_t = z_t \cdot c \). The spectrogram then becomes:

\[
E(f, z_t) = \left| \mathcal{F}(e(t)w(t - t_t)) \right| \quad (25)
\]

where \( \mathcal{F} \) denotes the absolute value of the Fourier transform (or frequency amplitude). Three features need to be set: the type and length of the window function and the level of overlap between two windows. The effect of the choice and type of window on the attenuation slope has been investigated (Akita and Ueda 1988; Kuc 1985); we extend this research to include the backscatter parameters as well.

Two experiments were carried out on a dataset of simulated RF signals under plane wave conditions, without attenuation, without diffraction, and using point scatterers. A Gaussian-shaped 3.75-MHz, 1.87-MHz bandwidth (–6 dB) pulse spectrum was used and the scatterer density within the pulse length was set at 10 scatterers, resulting in Rayleigh-distributed amplitudes, in which the condition defined in eqn (8) was shown to be true. One hundred acquisitions with 80 RF lines were generated and the four parameters were calculated as previously described using either a rectangular window, or a Hanning window.

In experiment 1, the window length was varied from 32 to 256 samples, with nonoverlapping windows. Figure 4 shows the STD and bias on the estimates. First, it is observed that the rectangular window produces lower STD than the Hanning window. Second, the rectangular window STD ("O") coincides very well with the theoretical STD ("x"). Finally, the bias is very low for both types of windows.

It is known that using overlapping windows decreases STD compared to using nonoverlapping windows in the calculation of the power spectrum (which is comparable to attenuation and backscatter coefficients). From Welch (1967), it can be concluded that using windows overlapping by 50% results in minimized STD of the attenuation and backscatter coefficients. The effect on the STD of the slope and intercept estimated from these coefficients is unclear. A second experiment was carried out to study the effect overlap has on the STD of the slopes and intercepts calculated from the coefficients. Figure 5 shows that overlapping...
has little or no effect in case of rectangular windows. Furthermore, increased overlap decreases STD of the Hanning windows up until an overlap of 50%, where Hanning windows STDs become comparable to rectangular window STDs. From the two experiments we conclude that the effect of the spectrogram calculation on the STD of the parameters is negligible and STD is rather insensitive to the choice of windowing function, type and overlap.

The second factor, $f_{bo}$, concerns the averaging of spectrograms into one average spectrogram. Overlapping of beams effectively decreases the number of lines (Kuc 1985). We estimated the lateral correlation coefficient of our acquisition system and found a correlation distance of 2.5 lines. Thus, our effective number of lines used in eqn (12) decreases by a factor of 2.5, which amounts to multiplying the predicted STDs by $f_{bo} = \sqrt{2.5} = 1.58$.

The third factor, $f_{dc}$, is due to imperfections in correcting for the TC. Imperfect correction demonstrates itself as a depth dependency of the parameter estimates. If the depth is ignored, the effect of the dependency is an increase in STD. The factor $f_{dc}$ measures this increase.

The fourth and fifth factors, local ($f_{li}$) and global ($f_{gi}$) inhomogeneities, both reflect the physical properties of the backscattering medium. In in vivo liver measurements and even in phantom measurements, the measured STD was higher than expected. We think this effect is the result of inhomogeneous physical properties. The factor $f_{li}$ is the excess STD within one sample of a medium as compared with theory. The factor $f_{gi}$ also measures excess STD, but between different samples of a medium. In other words, the expected STD within one liver is $f_{li}$ times the theoretical value, whereas the STD measured over a number of livers is $f_{li} f_{gi}$ times the theoretical value.

To conclude this section, we have shown that the first factor (spectrogram calculation) can be disregarded. The theoretical STD, $\sigma_{x,th}$, of parameter $x$, should thus be multiplied by the remaining four inhomogeneity factors to arrive at a practical estimate of the STD:

$$\sigma_{x,prac} = f_{li} f_{gi} f_{dc} f_{bo} f_{x,th}$$

where $f_{x,bo} = 1.58$.

**TRANSDUCER CHARACTERISTICS ESTIMATION**

Until now, the effect of the transducer on the parameter estimation has been neglected, because data were simulated under plane wave conditions. For the
Fig. 4. Estimated STD and bias of the four parameters from 100 simulated RF acquisitions (80 line, 1024 samples) using the rectangular (O), or Hanning (+) window. The theoretical STD (x) was calculated; theoretical bias was zero. The window length was varied while using nonoverlapping windows.

theory to be tested on actual data, adequate transducer characteristics (TC) estimation has to take place. We used a Toshiba-T270A scanner using the 3.75-MHz PSF37-CT phased array transducer in single focus mode (F = 7.5 cm). As explained in the Introduction, RF signals are acquired without effects of manual gain and/or TGC control of the scanner. In the Theory section, the TC was defined as the product of $P^2(f)$, the acoustoelectric transfer function of the transducer, and $D^2(f,z)$, the diffraction spectrogram of the transducer.

As explained in the Theory section, the diffraction could be medium-dependent. We therefore set out an experiment to investigate the influence of the medium on the TC estimation. We used three different media: foam, rubber and liver. The foam medium [also used by Lerski (1982)] is a 90-ppi (90 pores per inch; 3.54 pores per millimeter) reticulated polyurethane cellular network which is entirely open (Bulpren S90, Recticel, Kesteren, The Netherlands). The foam is immersed in water and thoroughly degassed using a (0.2-mbar end pressure) vacuum pump (S1.5, Leybold AG, Köln, Germany). The rubber medium is a Model 539 multi-purpose phantom (ATS Laboratories, Bridgeport, CT, USA). The rubber phantom contained test structures for quality assurance purposes. The condition defined in eqn (8) was verified to hold for both phantoms. In vivo liver data were acquired from seven male and five female normals, examined at 9.00 h before breakfast. At least four lateral and four transverse acquisitions in the right liver lobe were made, while avoiding regions with high concentrations of blood vessels. Two methods of TC estimation were used: (1) absolute and (2) relative. The foam TC was measured using an absolute method, and the rubber and liver TC were estimated using a relative method.

The absolute TC (on foam) was estimated using a method well known in the literature (Insana et al. 1983; Oosterveld et al. 1991). The procedure is as
follows: Place a volume of the medium in a water tank and orient its planar surface perpendicular to the beam axis. Record 1200 echo signals from adjacent, non-overlapping lines of sight. In each line, calculate the amplitude spectrum of a 128-point segment just beneath the surface, and average over all 1200 lines. Repeat this over a range of transducer–foam distances. The averaged spectra are then stacked into one, average spectrogram. The estimated diffraction spectrogram, $\hat{D}_{\text{abs}}(f, z)$, is obtained by dividing the average spectrogram by its focus spectrum, which is the maximum energy spectrum. Figure 6 shows the diffraction spectrogram; notice that it is flat in the focus (because of the division). Next, the acoustoelectric spectrum, $P_{\text{abs}}(f)$, should be estimated from a flat plate reflection at the focus (Nicholas et al. 1982; Ueda and Ozawa 1985). We were not able to record a reliable flat plate reflection due to multiple reflections caused by previously emitted pulses, and overflow due to the high reflectivity of the flat plate. Therefore, $P_{\text{abs}}(f)$ was measured indirectly. The backscatter coefficient of the foam was measured in a laboratory setup (Ueda and Ozawa 1985). The foam focus spectrum divided by the laboratory estimated backscatter coefficient is an indirect estimate of $P_{\text{abs}}(f)$. Figure 7 shows this pulse spectrum. The above method estimates TC in an absolute sense; i.e., the correction by an absolute TC results in absolute attenuation and backscatter measurements. The foam TC was measured independently from foam acquisitions used for parameter measurements.

The relative TC (on rubber and liver) was estimated using the following procedure. First, a maximum number of independent acquisitions is made on the medium. Next, the homogeneous STFT windows of all the lines in all acquisitions are averaged (>900 windows in rubber, and >5000 in human liver). The result equals the average spectrogram of the medium. In the same manner as above, the estimated diffraction

---

### Table 1: Estimated STD and Bias of Parameters

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Rectangular Window</th>
<th>Hanning Window</th>
</tr>
</thead>
<tbody>
<tr>
<td>STD $a_0$</td>
<td><img src="image1" alt="STD a0" /></td>
<td><img src="image2" alt="STD a0" /></td>
</tr>
<tr>
<td>Bias $a_0$</td>
<td><img src="image3" alt="Bias a0" /></td>
<td><img src="image4" alt="Bias a0" /></td>
</tr>
<tr>
<td>STD $a_1$</td>
<td><img src="image5" alt="STD a1" /></td>
<td><img src="image6" alt="STD a1" /></td>
</tr>
<tr>
<td>Bias $a_1$</td>
<td><img src="image7" alt="Bias a1" /></td>
<td><img src="image8" alt="Bias a1" /></td>
</tr>
<tr>
<td>STD $b_0$</td>
<td><img src="image9" alt="STD b0" /></td>
<td><img src="image10" alt="STD b0" /></td>
</tr>
<tr>
<td>Bias $b_0$</td>
<td><img src="image11" alt="Bias b0" /></td>
<td><img src="image12" alt="Bias b0" /></td>
</tr>
<tr>
<td>STD $b_1$</td>
<td><img src="image13" alt="STD b1" /></td>
<td><img src="image14" alt="STD b1" /></td>
</tr>
<tr>
<td>Bias $b_1$</td>
<td><img src="image15" alt="Bias b1" /></td>
<td><img src="image16" alt="Bias b1" /></td>
</tr>
</tbody>
</table>

Fig. 5. Estimated STD and bias of the four parameters from 100 RF acquisitions (simulated 80 line, 1024 samples) using the rectangular (○), or Hanning (+) window. Theoretical STD (x) was calculated; theoretical bias was zero. The level of overlap was varied while using 128-point windows.
spectrum, \( D_{\text{ad}}(f, z) \), is obtained by dividing the average spectrogram by its focus spectrum, which is the maximum energy spectrum. The acoustoelectric spectrum, \( \hat{P}_{\text{ae}}(f) \), is estimated by the focus spectrum. We had already assumed that the media were homogeneous, thus the average spectrogram will contain, besides the diffraction spectrogram and the acoustoelectric transfer function, also the average attenuation and backscatter coefficient. It follows that the resulting parameter measurements using the relative TC will be relative to the medium used. Thus, for normal liver measurements corrected with relative normal liver TC, the average estimated parameter should be zero. Interestingly, if the parameter is significantly (using STD) nonzero, then a ROI should be considered abnormal. Relative TC does not measure parameters in an absolute sense, but still has the diagnostic information at hand. Actually, the procedure to calculate the relative TC is a little more complicated, because the rubber, as well as the liver medium, contains nonhomogeneous tissue regions (e.g., blood vessels, reflectors). In the Parameter Estimation Method section, a manual outlining of organs as well as routines to detect the nonhomogeneous regions are available. This information was used in estimating the relative TC, by skipping windows that contained such a region.

As can be seen from Figs. 6 and 7, the TC estimates still show some noise, although each spectrum is the result of averaging 1200 spectra. TC correction not only removes the effect of the TC, but if a limited number of lines is used, the TC estimate itself is a noisy estimate [cf. eqn (12)] and will add noise as well, decreasing precision. To reduce the amount of noise introduced, the number of averaged RF lines in TC estimation should be high. This number depends on the amount of noise on LME expected in the ROIs when measuring parameters. The amount of noise in a ROI depends on the number of lines [eqn (12)].
Our maximal ROI size covers 110 lines. The number of lines in TC estimation should be much higher than the number of lines in a ROI. We took about 10 times the maximal number of lines in a ROI and found no influence on the precision of the estimated parameters.

Finally, two extra TCs are estimated. First, the relative normal human liver TC was estimated using all available normal human liver acquisitions of all subjects. In evaluating the efficacy of the TC correction, the same human liver material is used. Thus, a ROI in a liver being investigated is TC-corrected with data that contains information from that same liver. To avoid this so-called resubstitution effect, the leave-one-out (LOO) estimate of liver TC is also calculated. The LOO estimate is the TC estimation from all livers except the liver currently being investigated. In the experiments, it is indicated with “liver loo.” The second extra TC is a flat TC, to present an upper bound on TC correction error. It is a diffraction spectrogram and a pulse spectrum filled with ones, and represents the case where no diffraction correction is applied. In the experiments it is indicated with “none.”

RESULTS

It has been shown that four factors convert the theoretical STD estimates [eqns (21)–(24)] into practical estimates [eqn (26)]. The effect due to beam overlap was known in advance; in this section, the remaining three factors are estimated in two experiments. An explanation on how to estimate a factor is given in Appendix B.

Experiment 1

Parameters within the foam phantom were measured. Three types of materials were used for TC estimation: foam, rubber or a flat diffraction spectrogram. The results are shown in Table 1. The TC type is indicated in column 1; each was explained in the previous section. The type of inhomogeneity factor being measured is indicated in column 2 by mnemonics, which are explained in the caption. The following four columns show the inhomogeneity for each parameter. In between brackets, the 95% confidence interval of the point estimate is indicated.

It is observed that, for all TC types, the local inhomogeneity estimates are the same. This is to be expected as these inhomogeneities are estimated with fixed depth of the ROI. The local inhomogeneity is insignificant for the attenuation slope ($f_{a,1}$), but is significant for the other parameters. This indicates that the underlying physical parameters within the foam show local fluctuations.

The inhomogeneity due to diffraction correction ($f_{dc}$) varies with the type of TC. It is clear that foam-estimated TC results in the lowest ($f_{dc}$) values: both slope inhomogeneities are not significant, and both intercept parameters are smaller than the other TC types. From this experiment, it can be concluded that foam data cannot be corrected with a “rubber” calibrated TC, although both media are assumed to be tissue-mimicking media.

Finally, the remaining inhomogeneity was estimated. The whole dataset was transformed by subtracting one global average value, then each parameter was divided by the expected STD using eqn (26). The resulting inhomogeneity describes how well the model fits, and it is found not to be significantly different from 1. Thus, the STD model comprising the theoretical estimate times the various factors describes the data very well.

Experiment 2

Parameters within in vivo normal livers were measured in the same manner as with the phantom. Four types of TCs were now used: foam, rubber, none and

<table>
<thead>
<tr>
<th>TC</th>
<th>Type</th>
<th>$a_0$ (95% CI)</th>
<th>$a_1$ (95% CI)</th>
<th>$b_0$ (95% CI)</th>
<th>$b_1$ (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Foam</td>
<td>li</td>
<td>1.15 (1.10–1.19)</td>
<td>0.97 (0.93–1.00)</td>
<td>2.00 (1.92–2.07)</td>
<td>1.57 (1.51–1.63)</td>
</tr>
<tr>
<td></td>
<td>dc</td>
<td>1.15 (1.11–1.19)</td>
<td>1.03 (0.99–1.07)</td>
<td>1.07 (1.03–1.11)</td>
<td>1.02 (0.98–1.05)</td>
</tr>
<tr>
<td></td>
<td>mo</td>
<td>1.00 (0.97–1.04)</td>
<td>1.00 (0.96–1.04)</td>
<td>1.00 (0.97–1.04)</td>
<td>1.00 (0.96–1.03)</td>
</tr>
<tr>
<td>Rubber</td>
<td>li</td>
<td>1.14 (1.10–1.19)</td>
<td>0.97 (0.93–1.00)</td>
<td>1.99 (1.92–2.07)</td>
<td>1.57 (1.51–1.62)</td>
</tr>
<tr>
<td></td>
<td>dc</td>
<td>1.25 (1.20–1.29)</td>
<td>1.73 (1.67–1.79)</td>
<td>1.18 (1.13–1.22)</td>
<td>1.74 (1.68–1.81)</td>
</tr>
<tr>
<td></td>
<td>mo</td>
<td>1.00 (0.96–1.04)</td>
<td>1.00 (0.96–1.04)</td>
<td>1.00 (0.96–1.03)</td>
<td>1.00 (0.96–1.04)</td>
</tr>
<tr>
<td>Flat</td>
<td>li</td>
<td>1.14 (1.09–1.18)</td>
<td>0.96 (0.93–1.00)</td>
<td>1.97 (1.90–2.05)</td>
<td>1.58 (1.52–1.64)</td>
</tr>
<tr>
<td></td>
<td>dc</td>
<td>17.4 (16.7–18.0)</td>
<td>1.29 (1.25–1.34)</td>
<td>16.8 (16.2–17.5)</td>
<td>1.21 (1.17–1.26)</td>
</tr>
<tr>
<td></td>
<td>mo</td>
<td>1.00 (0.96–1.04)</td>
<td>1.00 (0.96–1.04)</td>
<td>1.00 (0.96–1.04)</td>
<td>1.00 (0.96–1.04)</td>
</tr>
</tbody>
</table>

Mnemonics: li is local inhomogeneity, dc is diffraction correction, and mo is model.
average human liver. The results are shown in Table 2. The global inhomogeneity measures the increase in STD when measuring different livers, whereas the local inhomogeneity measures increase due to inhomogeneity within the liver.

It is observed that, for all TC types, the local and global inhomogeneity estimates are the same. This is to be expected, as these are estimated with fixed depth of the ROI. The local inhomogeneity is very high for both attenuation intercept \((f_{\text{li},a})\) and backscatter intercept \((f_{\text{li},b})\). The local inhomogeneity is still high with respect to the other types of inhomogeneities for both attenuation slope \((f_{\text{li},a1})\) and backscatter intercept \((f_{\text{li},b1})\). The global inhomogeneity \((f_g)\) is much lower than the local inhomogeneity, and becomes nearly negligible for the slope values \((a1, b1)\).

The inhomogeneity due to diffraction correction \((f_d)\) varies with the type of TC. It is clear that the average liver type TC results in the lowest \((f_{dc})\) values. Neither of the backscatter parameters have significant inhomogeneity \((1 \text{ is included in the interval})\), and both attenuation parameters are nearly negligible. There is no difference between the leave-one-out (LOO) TC estimate and the resubstitution TC estimate. This indicates that the average liver TC predicts diffraction well in unseen livers.

Finally, the remaining inhomogeneity was estimated. The whole dataset (all livers, all sizes, all depths) was normalized by subtracting the global average value, and subsequently dividing by the STD which was obtained by multiplying the theoretical values with the estimated inhomogeneity factors and the beam overlap factor. The resulting inhomogeneity observed in all measurements is not significant. Thus, again, the STD model comprising the theoretical STD estimate times the various factors describes the data very well.

 Instead of looking at it quantitatively, Fig. 8 gives a qualitative view on the depth-dependent diffraction filter effect. Five sets of average parameters are plotted versus depth, from five types of TC corrections. To compare the sets of parameters, the mean values in each set were subtracted. It is clearly seen that the liver TC results in the smallest depth-dependency effect. Both rubber and foam result in the same type of errors.

**DISCUSSION**

We have shown that the STDs of four in vivo measured acoustospectrographic parameters are predicted very well. The predicted STD comprises: a theoretical part based on a homogeneous tissue model with Rayleigh-distributed backscattered amplitudes; and a practical part with various factors that account for deviations from the model.

Throughout this study, important concepts have emerged, and we will summarize them by following the processing steps.

For the parameter estimation methods to be clinically applicable, an essential feature is the possibility to
acquire accurately radio-frequency (RF) signals from standard clinical systems. These systems should have operator-independent RF signals available. We use a system expanded with a custom-built RF interface to avoid operator-dependent gain and/or TGC settings in the RF signal. Furthermore, an acquisition workstation must be available that can acquire these high-frequency signals over the wide dynamic range, related to the attenuation in the signals. Our 8-bit 50-MHz acquisition system workstation with a reproducible TGC amplifier (Verhoeven and Thijssen 1992), in combination with the RF interface, thus forms a firm basis for our methods.

The next step in robust estimation of parameters is to build in a strategy to account for in vivo measurement conditions. We use a mask matrix that is used to store manually and automatically detected areas such as blood vessels, ligaments, and over- and underflow of the digitizer. When calculating the average spectrogram, windows that contain any marked samples are left out of the analysis, as these windows could influence parameter estimates. The methods were verified by visual inspection, which revealed that all visible structures were properly detected. We have set the detection threshold such that it is biased toward detection, which assures that the remaining area is without these structures. In this article, we selected only ROIs with less than 5% masked area.

The number 5.57 dB cited in literature when calculating the expected STD of the log mean average spectrogram (Berger et al. 1987; Kuc and Schwartz 1979), pertains to calculating the mean of the logarithm of Rayleigh-distributed variables. We calculate the log of the mean and find 4.54 dB (eqn (12)). Figure 9 shows the results of simulations while using both calculation strategies, where STDs are multiplied by $\sqrt{N}$. The figure clearly shows that the two methods do not produce the same results. It can be concluded that the log of the mean results in better precision than the mean of the log when calculating the log average spectrogram.

A consistent combined analytic derivation of the STD of all four parameters [eqns (21), (22), (23) and (24)] has not yet been described in literature. The STD of the attenuation slope [fit through zero, i.e., $\alpha(f) = a_0 + a_1 \cdot (f - f_0)$] was analytically derived by Kuc (1985), Parker (1986) and Berger et al. (1987). We applied a two-parameter model $[i.e., \alpha(f) = a_0 + a_1 \cdot (f - f_0)]$ with fewer approximations, and used the log mean spectrogram. Simulations show that our equations predict the STD of the parameters measured from signals generated according to the signal model very well over

---

**Fig. 8.** *In vivo* liver, average acoustospectrographic parameters measured in regions-of-interest at various depths. Three TCs were used, calibrated on: foam or rubber (+); liver (8, resubstitution and leave one out); flat spectrogram, i.e., no diffraction correction (O).
large depth and frequency ranges. As has been recognized by Parker (1986), longer ROIs enable estimation of attenuation more accurately than wider ROIs, because averaging adjacent scan lines reduces the STD by only the square root of the number of lines, whereas the STD reduces by the ROI length to a power f. The backscatter parameter STD, however, is reduced equally by both increasing the number of lines as well as increasing the ROI depth. The equations show that the STDs are independent of absolute attenuation value, as was also noticed by Parker (1986); we extend this by recognizing that they are also independent of the absolute backscatter value.

An intermediate step in the methods is the calculation of the spectrogram from the RF signal. These calculations require that the length, degree of overlap, and type of windowing function be chosen in advance. We have shown that the rectangular window produces near theoretical STDs, and that a Hanning and rectangular window have equal performance when overlapping the windows by 50%. Furthermore, simulated results show that STD and bias are not sensitive to modest variations in window size and overlap. For all windows, over a wide range of window sizes and overlap, the bias on the parameters is very low. The latter two observations thus alleviate careful tuning strategies of the three settings.

So far, results were shown on simulated data; when introducing the transducer with its typical characteristics (TC) on the received backscatter spectrum, a correction has to be performed. We have shown two strategies to measure the TC. From the results, we conclude that TC correction varies with the type of medium as was also noticed by Robinson et al. (1984). The effect of TC correction on parameters measured on in vivo liver data is very small only when TC is calibrated with in vivo liver data. We stress that our parameters are measured on unseen liver data in the calibration (as observed in the leave-one-out experiment). Foam, rubber phantoms, as well as no TC correction, all show significant influence of TC on parameter estimates in the liver.

We measured two types of inhomogeneity due to fluctuations of underlying physical parameters: local (within an organ), and global (between organs, or intersubject). We were surprised to find that even rather homogeneous phantom material shows significant local inhomogeneity. Our results for in vivo data show that local inhomogeneity is the major factor in increasing STD estimates with respect to theory. The increase due to global variations is much smaller. Therefore, we think that any research in improving results on parameter estimation should focus on the local inhomogeneity.

We have estimated factors describing the increase in STD on normal in vivo liver data. We have shown that we can predict the STD on in vivo data very well. We propose a new acoustic parameter: the local inhomogeneity. We have shown a method to calculate the local inhomogeneity in normal human liver. If we examine a diseased organ, it might be anticipated that the local inhomogeneity could also change due to changed tissue composition. Inhomogeneities indicated in this article could be compared with measured inhomogeneities, if they show significant deviation; it could then be concluded that the organ under investigation is abnormal with respect to local tissue inhomogeneity.

Acknowledgements—The authors thank the referees for their helpful comments.

REFERENCES


**APPENDIX A: DERIVATION OF EQNS (13) AND (14)**

In this appendix, eqns (13) and (14) are derived. Given a set of measurements \(y_i, x_i\) which were generated by a linear model:

\[ y = ax + b + \epsilon \]

If \(\epsilon\) is an independent, zero mean, additive and Gaussian-distributed stochastic variable, \(N(0, \sigma)\), then the variance in the estimated slope and intercept is (Kleinbaum et al. 1988):

\[ \sigma_y^2 = \frac{\sum (y_i - \bar{y})^2}{N} \]

\[ \sigma_x^2 = \frac{\sigma^2 \sum x_i^2}{N \sum (x_i - \bar{x})^2} \]

In the current application, the \(x_i\) are sampled at equidistant intervals \((x_{i+1} - x_i = \Delta x)\), which enables the summations to be developed into simple expressions. Equation (28) becomes:

\[ \sigma_x^2 = \frac{12\sigma^2}{\Delta x^2(N - N)} \]

which is equivalent to the result by Berger et al. [1987, eqn (5)].

For the development of eqn (29), an extra term is introduced, \(M = x_i/\Delta x\), which is the number of intervals delay from 0 to \(x_i\). Equation (29) then becomes:

\[ \sigma_x^2 = \frac{2\sigma^2 N}{(2N^2 + 6N + 6M^2 - 3N - 6M + 1)} \]

\[ \frac{N(N - N)}{N - N} \]

It should be noticed that eqns (30), and (31) are exact expressions of eqns (28) and (29). They are more accurate than the approximations at this stage by Parker [1986, eqn (22)], and Yao [1991, eqn (12)].

Equations (30) and (31) can be approximated if it is assumed that \(N > 2\). Then, \(N^2 - N \approx N(N - 1)\) and \((2N^2 + 6N + 6M^2 - 3N - 6M + 1) \approx (2N^2 + 6M + 6M^2 - 3N - 6M)\). Substituting the total length of a ROI, \(D = \Delta x N\), and the relative delay, \(\delta = M/N\), results in the following approximations of eqns (30) and (31):
\[ \sigma_x^2 = \frac{4\sigma^2}{D^3N} \quad (32) \]
\[ \sigma_x^2 = \frac{4\sigma^2 \left(1 + 36 + 3\delta^2 - \frac{3 + 6\delta}{2N}\right)}{N} \quad (33) \]

Comparing eqns (7) and (27) it is seen that the attenuation equals \( a/2 \), thus eqn (32) should be divided by 4. The square root then results in eqns (13) and (14).

**APPENDIX B: ESTIMATING INHOMOGENEITIES**

A factor, \( f \), represents the increase in estimated STD, \( \delta \), relative to the expected STD, \( \sigma \), due to variations in an accompanying setting, \( F \), thus \( \delta = f\sigma \). For example, the factor due to incorrect TC correction is 1 if the depth is fixed. In this single factor model, \( f \) is simply estimated by calculating \( \delta \) from the whole dataset, and dividing by \( \sigma \), which is calculated from theory. For our acoustic parameter STDs this is not possible, because the setting \( F \) also influences the expected STD; thus \( \delta(F) = f\sigma(F) \). As an example, consider ROI parameters calculated at different depths, as shown in theory, the expected STD varies with depth.

To explain how the factor can then be calculated, let us assume that an acoustic parameter, \( \alpha \), is a stochastic variable with the following probability density function (pdf) with STD dependent on setting \( F \):

\[ p_1(x(F)) = N(x, \sigma(F)) \quad (34) \]

Comparing eqns (7) and (27) it is seen that the attenuation equals \( a/2 \), thus eqn (32) should be divided by 4. The square root then results in eqns (13) and (14).

The normality assumption \( (N(.,.)) \) was already shown in the subsection, STD of the Attenuation and Backscatter Slope and Intercept. The average of the parameter is constant \( (\bar{x}) \) and should not depend on \( F \) (at least, that is the hypothesis). Transforming the parameter \( (x') \) by subtracting the mean (calculated over the whole dataset) and then dividing it by the expected STD results in a new stochastic variable with the following pdf:

\[ p_2(x') = p_0 \left( \frac{x(F) - \bar{x}}{\sigma(F)} \right) = N(0, f) \quad (35) \]

The factor \( f \) is now simply estimated by calculating the STD from the transformed variable \( (f = \sigma_{x'}) \).

Instead of one, three factors have to be estimated. Then, a single factor is estimated by calculating the average while keeping the other settings constant; e.g., using \( x_{r1}, r_3(F_2) \) estimates \( f_2 \).