1 Introduction

Ultrasound characterisation of the structure of soft biological tissues is based on the scattering from its complex inhomogeneities which mainly include proteins and lipids. To characterise these, several empirical formulations relating the ultrasonic attenuation, velocity and scattering data, and biochemical estimates have been reported (O'BRIEN, 1977; FREESE and LYONS, 1979). In addition to these, the root mean square of ultrasonic velocity fluctuations $\mu$ and maximum spacing or size of scatterer $d_a$ are employed to describe quantitatively the changes in tissue structure (SEHGAL and GREENLEAF, 1984). For normal soft tissues, a $\mu$ value around 2.73 per cent represents the compressibility variations (CRAVERS, 1977). The change in this parameter could indicate the tissue changes both at the macroscopic (mm) and microscopic (few tens of $\mu$m) levels (NICHOLAS, 1982a).

The studies carried out by CHIVERS et al. (1974) and LIZZI and ELBAUM (1979) show that the structural variations in tissues influence the backscattering amplitude ($BSA$) and its frequency ($f$) dependence (hereafter referred to as $BSA(f)$), which is mainly due to large concentrations of small sized scatterers and large interfaces which, respectively, scatter isotropically and reflect ultrasound specularly. In normal tissues, the large specular interfaces are randomly oriented to the ultrasonic beam and thus their spatial averaging leads to weak but deterministic modulation in $BSA(f)$. These modulations, recovered by signal-processing techniques, are employed to determine the maximum scatterer size $d_a$ (FELLINGHAM and SOMMER, 1984). However, the macrostructure, which represents a distribution of scatterer size $d_j$, is better described by a distribution of a spacing function $F(d_j)$.

In contrast to this method, the tissue structure at the microlevel is described by the corresponding data of $\mu$ and $d_a$, obtained by curve fitting of the experimental data of the frequency dependence of the backscattering coefficient $\eta(f)$ (SEHGAL and GREENLEAF, 1984). To obtain these parameters an appropriate choice of filters and optimum gate widths are employed and are compared with the known values of a sponge sample (polyurethane foam, density 20 kg m$^{-3}$) (NICHOLAS, 1982b).

The aim of the present study is to apply the above techniques to obtain the data of following tissues, which encompass various pathological structures:

(a) hypercholesterolemic liver (rabbit)
(b) lymphoid-lucosed liver (hen)
(c) rhinosporidium tissue (human)
(d) ethmoidal tumour (bovine).

The data on these tissues, which have not been available so far, may help the understanding of the growth processes of these disorders in various species.

2 Materials and method

2.1 Tissue macrostructure parameters

To obtain the spacing distribution function $F(d_j)$ the following approach, a modified version of the method of FELLINGHAM and SOMMER (1984), has been utilised. It is based on the following assumptions.

(a) The scatterers are arranged in an inhomogeneous array in some characteristic pattern on a scale comparable to the acoustical wavelength.
(b) The scattering is weak due to the random nature of the orientation of the reflecting interfaces and is contributed mainly by the elastic variations of the interfacial components such as collagen. Density variations are neglected.
(c) The tissue is assumed to contain scatterers with spacings $d_j$, where $j = 1, 2, \ldots, n$. $d_j$ is given in mm.

The analytical process (Fig. 4) is carried out in the following steps:

(i) Spectral subtraction to obtain $(f_s - f_0)$ where

$$f_s = \log (BSA_{s}(f))$$

and

$$f_0 = \log (BSA_{0}(f))$$

(ii) Weighting by Hann window to remove noise introduced at the lower and upper end of the frequency spectrum displayed in the time window of a swept frequency spectrum analyser.

(iii) Differentiation of $H(f_s - f_0)$ to obtain $H' = H'(f_s - f_0)$. This step improves the high-frequency components and gives an amplitude weighting of $A_m$ which is proportional to the frequency $f_{m,j}$ of the fluctuations.

(iv) Autocorrelation of $H' = R(H') \equiv R$. This process brings out the weak but deterministic components of $H'$, which are related to the structure spacing $d_j$.

(v) Spectral analysis of $R$ to obtain $F(R) = F'(d_j)$ setting bounds $(d_1, d_2)$ for $d_j$ as $d_1 < d_j < d_2$.

For a sweep rate of 10 MHz/2 s of the analyser, the spacing 0-15 mm corresponds to 1 Hz in $f_{m,j}$. For a rectangular gate width of $\tau_\varphi = 5\mu s$ the upper limit of $d_s$ occurs as an artefact at 25 Hz = 3-75 mm, corresponding to the spatial extent of gate. The lower limit $d_1 = 0-45$ mm.

2.2 Tissue microstructure parameter

According to the theory of Sehgal and Greenleaf (1984) the ultrasonic backscattering coefficient $\eta(f)$ is given by

$$\eta(f) = \frac{3\mu^2k}{4} \left[ \arctan \frac{kd_m}{(1 + k^2d_m^2)} + \frac{kd_m}{3(1 + 9k^2d_m^2)} \right]$$

(1)

where

$$k = 2\pi f/c_s$$

and $\mu^2 = \text{mean square fluctuation in ultrasonic velocity}$

Here $\eta(f)$ is determined experimentally and values of $d_m$ and $\mu^2$ are determined by data fitting. Eqn. 1 shows that the frequency dependence varies from 1 to 4 as $kd_m$ varies from macroscopic to microscopic situations.

Experimental measurement of $\eta(f)$ is carried out by a substitution technique (O'Donnell et al., 1981) by substituting various parameters (Fig. 1) in the formula:

$$\eta(f) = \frac{r_x(s(f)) R F}{2T} \text{cm}^{-1} \text{sr}^{-1}$$

(2)

where

$R = \text{axial range of scattering volume } v_x(f)$

$r_x(f) = \text{scattering volume computed at frequency } f$

$r_x = \text{ultrasonic reflection coefficient between the plane reflector and bath}$

$T = \text{ultrasonic transmission coefficient between the bath and the sample}$

$s(f) = \text{backscatter transfer function of sample independent of transducer system, estimated from spectral analysis of scattered echoes from sample and reflection echo at the same range.}$

It is given by

$$s(f) = 10^{-2S_{ref}(f) - S_{ref}(f)/10}$$

(3)

Fig. 1 Scattering geometry showing different parameters such as gated volume $v_x$, gate width $\tau_\varphi$, beam profile diameter $\phi(R, f)$ at range $R$ and frequency $f$.

where

$S_{ref}(f) = \text{spectral amplitude of the reflector echo measured in water, dB}$

$S_x(f) = \text{BSA}(f) \text{ from sample, dB}$

$F(x, f), \tau_0, \tau_\varphi = \text{function representing attenuation loss in the part of tissue between the transducer and scattering volume (F), given by}$

$$F(x, f), \tau_0, \tau_\varphi = \exp \left( \frac{\tau_0 x_0}{(2\tau_0 x_0)} \right)$$

(4)

where

$\tau_0 = \text{gate length, s}$

$x_0 = \text{axial depth of centre of scattering volume from tissue surface}$

$\alpha(f) = \text{attenuation coefficient in tissue as a function of } f, \text{np cm}^{-1}$

2.3 Description of tissue samples

2.3.1 Hypercholesterolemic (HC) rabbit liver. The histological picture of the liver reveals (Fig. 2a) congestion of central veins. The hepatic cells show hydropic degenerative changes (Jones and Hunt, 1983).

2.3.2 Lymphoid lucosis (LL) of hen liver. Lymphoid lucosis occurs in birds. It is a lymphoma tumour affecting all visceral organs mostly involving the liver, also commonly known as 'big liver' disease. The liver is grossly enlarged 3-4 times its normal size. The histological appearance of such affected liver shows (Fig. 2b) massive infiltration of lymphoblastic series of neoplastic cells replacing hepatic cells. The centre of the field reveals invasion of the hepatic cells whereas its periphery shows degenerating hepatic cells (Gordon and Jordan, 1982).