Measurement of breast-tissue x-ray attenuation by spectral mammography: solid lesions

Erik Fredenberg¹, Fleur Kilburn-Toppin², Paula Willsher², Elin Moa¹, Mats Danielsson⁴, David R Dance⁵,⁶, Kenneth C Young⁵,⁶ and Matthew G Wallis²,³

¹ Philips Health Systems, Mammography Solutions, Smidesvägen 5, 171 41 Solna, Sweden
² Cambridge Breast Unit, Addenbrookes Hospital, Hills Road, Cambridge CB2 0QQ, UK
³ NIHR Cambridge Biomedical Research Centre, Addenbrookes Hospital, Hills Road, Cambridge CB2 0QQ, UK
⁴ Department of Physics, Royal Institute of Technology (KTH), AlbaNova University Center, 106 91 Stockholm, Sweden
⁵ NCCPM, Royal Surrey County Hospital, Guildford GU2 7XX, UK
⁶ Department of Physics, University of Surrey, Guildford GU2 7XH, UK

E-mail: erik.fredenberg@philips.com

Received 13 October 2015, revised 14 December 2015
Accepted for publication 2 February 2016
Published 10 March 2016

Abstract
Knowledge of x-ray attenuation is essential for developing and evaluating x-ray imaging technologies. For instance, techniques to distinguish between cysts and solid tumours at mammography screening would be highly desirable to reduce recalls, but the development requires knowledge of the x-ray attenuation for cysts and tumours. We have previously measured the attenuation of cyst fluid using photon-counting spectral mammography. Data on x-ray attenuation for solid breast lesions are available in the literature, but cover a relatively wide range, likely caused by natural spread between samples, random measurement errors, and different experimental conditions. In this study, we have adapted a previously developed spectral method to measure the linear attenuation of solid breast lesions. A total of 56 malignant and 5 benign lesions were included in the study. The samples were placed in a holder that allowed for thickness measurement. Spectral (energy-resolved) images of the samples were acquired and the image signal was mapped to equivalent thicknesses of two known reference materials, which can be used to derive the x-ray attenuation as a function of energy. The spread in equivalent material thicknesses was relatively large between samples, which is likely to be caused...
mainly by natural variation and only to a minor extent by random measurement
errors and sample inhomogeneity. No significant difference in attenuation was
found between benign and malignant solid lesions. The separation between
cyst-fluid and tumour attenuation was, however, significant, which suggests it
may be possible to distinguish cystic from solid breast lesions, and the results
lay the groundwork for a clinical trial. In addition, the study adds a relatively
large sample set to the published data and may contribute to a reduction in the
overall uncertainty in the literature.

Keywords: x-ray imaging, mammography, spectral imaging, photon
counting, tissue, x-ray attenuation

(Some figures may appear in colour only in the online journal)

1. Introduction

Basic knowledge of the x-ray attenuation of tissue is essential for the development of new
x-ray imaging technologies, as well as the study of the performance of existing technologies.
Knowledge of the x-ray attenuation of tissue is particularly important for the development of new
applications of unenhanced spectral imaging. Unenhanced spectral imaging is an emerging x-ray
imaging technology that measures tissue properties, without injection of a contrast agent, using
differences in attenuation as a function of energy. In the field of mammography, unenhanced
spectral imaging has been employed to improve the image signal-to-noise ratio (Berglund et al
2014), improve lesion visibility (Johns and Yaffe 1985, Taibi et al 2003, Kappadath and Shaw
2008, Fredenberg et al 2010b), and to measure breast density (Ding and Molloi 2012).

Another potential application of unenhanced spectral imaging is to distinguish cysts from
tumours and thereby address the relatively low specificity of x-ray mammography screening
(Norell et al 2012, Erhard et al 2015). Round lesions are a common mammographic finding,
and it can be difficult to distinguish benign cysts from tumours on mammography alone, particu-
larly when the margin is partly obscured. Recall rates for assessment are approximately
5% in Northern Europe and much higher in the United States (Smith-Bindman et al 2003).
Not only is this costly for the screening programme (Guerriero et al 2011), but recalls can
be stressful for patients (Brett and Austoker 2001). The evidence that women may be put off
returning for subsequent screens following false positive recalls is mixed (Román et al 2011,
Brewer et al 2007, Maxwell et al 2013), but this potentially reduces the long-term effectiv-
ness of the screening programme. Improving lesion characterisation at screening would be
very desirable to reduce recalls, but the development of spectral x-ray techniques for this
purpose has been hindered by a lack of tissue attenuation data.

A few studies have reported measurements of the linear attenuation coefficient of solid
breast lesions, mainly with the purpose of comparing to normal fibro-glandular tissue.
Pioneering work was done by Johns and Yaffe (1987) who measured the linear attenuation
coefficient (energy range 18 to 110 keV) of 6 samples of infiltrating ductal carcinoma using a
broad x-ray spectrum and a spectroscopy detector. It was concluded that the attenuation of car-
cinoma was significantly different from that of fibro-glandular breast tissue at x-ray energies
below 40 keV. Carroll et al (1994) measured the linear attenuation coefficient (energy range
14 to 18 keV) of 12 samples of malignant breast tumors using monochromatic synchrotron
radiation. The study reported a difference in mean values between normal and cancerous
tissue, but there was overlap of the distributions. Chen et al (2010) measured the linear attenu-
ation coefficient (energy range 15 to 26.5 keV) of ≤14 samples (not all samples were imaged
at all energies) of mainly ductal carcinoma using computed tomography with monochromatic synchrotron radiation. The study did not find a significant difference between cancerous and fibro-glandular tissue. Tomal et al (2010) measured the linear attenuation coefficient (energy range 8 to 30 keV) of 18 invasive ductal carcinomas and 6 fibroadenomas using an x-ray tube and a silicon monochromator. A significant difference in attenuation between normal and cancerous tissue was found below 28 keV.

We have previously developed a method to measure the energy-dependent x-ray attenuation of tissue samples using spectral imaging (Fredenberg et al 2013). The method is based on mapping of the attenuation to equivalent thicknesses of two reference materials. A photon-counting spectral mammography system was used together with the method to measure the attenuation of cyst fluid, which marked the first step in our efforts to evaluate the feasibility of using unenhanced spectral imaging to distinguish between cysts and tumours in screening images. The method was validated by comparing measurements with calculations of attenuation from the measured elemental composition of cyst fluid and the known elemental composition of water.

The purpose of the current study is to measure the attenuation of formalin-fixed solid breast lesion specimens with the same methodology as for cyst fluid, to establish whether the attenuation of solid lesions is different enough from the attenuation of cystic lesions so that the two lesion types could be distinguished from one another by spectral imaging. Such a comparison is difficult to do with published data based on a different measurement setup because the potential bias from the different setups might obscure the small attenuation differences between the tissue types. However, data on tissue attenuation may also have general value for the scientific community as the sources of such data are sparse.

2. Materials and methods

2.1. Spectral mammography system

The Philips MicroDose SI mammography system comprises a tungsten-target x-ray tube with aluminium filtration, a pre-collimator, and an image receptor, which is scanned across the object (figure 1, left). The image receptor consists of photon-counting silicon strip detectors with corresponding slits in the pre-collimator (figure 1, right). This multi-slit geometry rejects virtually all scattered radiation (Åslund et al 2006). 32kV acceleration voltage was used for all measurements presented in this study and the exposure was set to 40 mAs per image. The relatively high exposure level was used in order to minimize quantum noise and resulted in an entrance surface air kerma of 6.7 mGy.

Photons that interact in the detector are converted to pulses with amplitude proportional to the photon energy. A low-energy threshold rejects virtually all pulses below a few keV, which are considered to be generated by noise. An additional high-energy threshold sorts the detected pulses into two bins according to energy. The high-energy threshold was set close to 20 keV, which yields approximately equal number of counts in each bin for a typical transmitted spectrum. The energy resolution at this energy level is approximately 5 keV full width at half maximum (Fredenberg et al 2010a).

2.2. Background of spectral imaging

X-ray attenuation at mammographic x-ray energies is approximately made up of only two independent interaction effects, namely photoelectric absorption and scattering processes
(Alvarez and Macovski 1976, Lehmann et al 1981, Johns and Yaffe 1985). Accordingly, a linear combination of any two materials of different and low atomic number can approximately simulate the energy-dependent attenuation of a third material of a given thickness,

\[ t_{\text{sample}} = t_1 \mu_1(E) + t_2 \mu_2(E) \]

We call these materials reference materials, and if this relationship is assumed to hold exactly, then the associated normalized reference thicknesses \( t_{\text{normalized}} = t_{\text{sample}} \) are unique descriptors of the energy dependent sample attenuation \( \mu_{\text{sample}} \) given the known attenuations of the reference materials \( \mu_1 \) and \( \mu_2 \). In other words, the detected signal in an x-ray detector would be identical for a tissue sample and for the equivalent combination of reference materials, regardless of incident energy spectrum or detector response.

We have previously developed a method to measure the energy-dependent x-ray attenuation of tissue samples that builds on equation (1) (Fredenberg et al 2013). In summary, aluminium (Al) and polymethyl methacrylate (PMMA) were used as the two reference materials, and a step wedge made of these two materials was positioned adjacent to the sample to provide a reference grid of thickness/material combinations for comparison to the sample. X-ray attenuation was measured by mapping the high- and low-energy counts obtained from a region-of-interest (ROI) located on the sample against those obtained from ROIs on the step wedge. Linear Delaunay triangulation in the log domain was used to find intermediate reference values. The method assumes that scattering processes can be treated as absorption, which is true only for x-ray detector geometries with efficient scatter rejection, such as multi-slit (Aslund et al 2006).

In common with previous studies (Lehmann et al 1981, Johns and Yaffe 1987), it is useful to express the equivalent Al and PMMA thicknesses in terms of the corresponding Al-PMMA vector with magnitude and angle given by
The magnitude \( r \) is directly proportional to the thickness and the density (specific weight) of the sample, whereas the angle \( \theta \) is related to the attenuation energy dependence and the (effective) atomic number of the material, and is independent of sample thickness.

In order to discuss our results in relation to other measurements of tissue attenuation, we have converted values of the linear attenuation reported in the literature to equivalent material thicknesses using the procedure described by Fredenberg \textit{et al} (2013). In summary, \( t_1 \) and \( t_2 \) were determined by the following least-squares fit to the published linear attenuation \( (\mu_{\text{sample}}) \) over the x-ray spectrum energy bins \( (E_1 \ldots E_N) \):

\[
\min_{t_1,t_2} \sum_{n=1}^{N} \phi(E_n) \Gamma(E_n) [t_1 \mu_1(E_n) + t_2 \mu_2(E_n) - \mu_{\text{sample}}(E_n)]^2.
\]

The fit was weighted by the expected spectrum after the sample \( (\phi(E)) \), calculated from a generic tungsten spectrum model (Boone \textit{et al} 1997), attenuated by 0.5 mm Al and the appropriate amount of PMMA from the sample holder, and the quantum efficiency of a 3-mm-thick silicon detector \( (\Gamma(E)) \).

2.3. Spectral attenuation measurements

In total, 84 formalin-fixed tissue samples of solid benign and malignant breast lesions were included in the study. Ethical approval was obtained to use samples from women from whom generic consent had been obtained prior to surgery. The generic consent allowed tissue to be used for ethically approved research, including genetic profiling. Immediately post-surgery the tissue was sliced to facilitate even fixation with formalin. For inclusion in this study, sliced samples were obtained from pathology the morning after surgery. Only samples that had the solid breast lesion clearly visible on both surfaces of the sample slice were used to avoid interference from normal breast tissue.

The samples were placed in a hollow PMMA cylinder with a threaded lid, henceforth referred to as the sample holder. By twisting the lid, the sample was gently compressed and the air gap between sample and lid was removed. A thin non-revolving PMMA plate between the sample and the threaded lid allowed compression without twisting or otherwise damaging the sample. Figure 2 illustrates the measurement setup.

The sample thickness \( (t_{\text{sample}}) \) was measured using two methods: (1) A protractor on the threaded lid allowed thickness measurement with a resolution of approximately 10 \( \mu \text{m} \) by calibrating the protractor on a series of gauge blocks, and (2) the height of the cylinder was measured with a calliper, also with a resolution of 10 \( \mu \text{m} \), from which the known bottom and lid thicknesses were subtracted. The means of the two thickness measurements were used in the calculation to produce normalized values of \( t_{\text{Al}} \) and \( t_{\text{PMMA}} \).

The samples were imaged with the mammography stand in horizontal mode. ROI selection was done by a medical physicist and verified / adjusted by an experienced radiologist. Care was taken to avoid areas with visible microcalcifications. An Al and PMMA step wedge was positioned adjacent to the sample in each image, and the range of the step wedge was adapted to the sample thickness by adding thin PMMA plates with an Al sheet on top, henceforth referred to as step-wedge plates. For each sample, the appropriate number of step-wedge plates was added so that the signal in the sample ROI fell within the range of signals from the ROIs on the step wedge. The thickness of PMMA in the reference grid was calculated as the
The difference between the total thickness of PMMA at the step wedge (including the step-wedge plates, step wedge and mounting plates) and the total thickness of PMMA at the sample holder (including all sample holder components and the mounting plates). The thickness of Al in the reference grid was calculated as the total thickness of Al at the step wedge (including the step-wedge plates and step wedge). The increased traversed thickness at oblique incidence was taken into account for calculating thicknesses in the reference grids.

2.4. Systematic and random measurement errors from thickness and density uncertainties

The thicknesses of the PMMA step wedge and the sample-holder components were measured with a micrometre screw, and the PMMA density was determined by weighing the step wedge and pieces of sample holder material with high-precision scales and measuring their size with the micrometre screw. The thickness of the step-wedge plates was measured with a micrometre screw, but the density was not measured and instead assumed to be equal to the average of the measured densities. The thicknesses of the PMMA mounting plates were measured with a calliper in a grid of positions at the step wedge and sample locations so that variations in the thickness could be taken into account. The PMMA densities of the mounting plates were not measured and again assumed to be equal to the average of the measured densities because the difference in thickness between the sample and step-wedge locations is small and any reasonable deviation from a standard value would therefore have minimal impact.

Measurement errors caused by thickness uncertainties in the measurement setup were divided into systematic and random errors, where the systematic errors are constant (related to measurement accuracy) and the random errors vary from sample to sample (related to measurement precision). Systematic errors may be caused by uncertainties in thickness and density of the reference materials, whereas random errors can be expected from variations in Al and PMMA thickness over the measurement area (considered random because the ROI position varied between measurements) and uncertainty in the sample thickness measurements.

Random errors in the PMMA thicknesses of all components were estimated as the standard deviation of measurements at several locations, or as the resolution of the measurement device, whichever was largest. A systematic error in PMMA thicknesses corresponding to
the resolution of the measurement device (calliper or micrometre screw) was assumed for each component. Systematic PMMA density errors were found by propagating weighing and volume errors (estimated by repeated measurements) through the density calculation. For components without measured density (the step-wedge plates and the mounting plates) a systematic error was estimated as the approximate variation of densities in off-the-shelf PMMA from different manufacturers.

The Al steps in the step wedge were constructed from layers of Al foil (purity 99.999%). The thicknesses of these were determined by measuring the area and weight of an approximately 100 \(\times\) 100 mm\(^2\) Al sheet, assuming the elemental density, and solving for the thickness. A systematic thickness error was estimated by propagating the weighing and area errors through the calculation. The weighing error was estimated by repeated measurements and the area error was estimated by the resolution of the measurement device (a ruler). The Al sheets on the step-wedge plates were made of the same type of foil and the elemental density was assumed, but the thickness was instead measured with a micrometre screw and a systematic error was estimated as the resolution of the measurement device. Random errors in Al thickness for the step wedge and step-wedge plates were estimated as the standard deviation of measurements with a micrometre screw at several locations on the Al sheets.

As the sample thickness measurement was repeated for each individual sample, i.e. the sample thickness was not constant, we can expect a random uncertainty in thickness, which shows up as a scaling of \(r\) (see equation (2)). These random variations were estimated as the standard deviation of the difference between the two thickness measurements (protractor and calliper) for all samples. Note, however, that this error estimation does not capture potential air gaps between the lid and the sample (the samples were not re-compressed between the two measurements), but we assume this source of error to be negligible. The resolution of the protractor and calliper was included as a systematic error.

2.5. Measurement variabilities

Each manually selected ROI was automatically divided into four equal-sized sub ROIs in order to estimate inhomogeneities within an ROI (denoted \(\sigma_{\text{ROI}}\)), and four images with identical ROI selection were acquired of each sample to estimate random fluctuations between measurements (\(\sigma_{\text{drift}}\)). The variation between the sub ROIs in each image, averaged over all images of the sample, is referred to as the intra-image variability (\(\sigma_{\text{intra}}\)). The variation between sub ROIs with equal position but in different images, averaged over all sub ROI positions, is referred to as the inter-image variability (\(\sigma_{\text{inter}}\)). The quantum-noise contribution to the variability of each sub ROI in each image (\(\sigma_{\text{quant}}\)) was estimated from knowledge of the photon counts per pixel in the high- and low-energy images (directly proportional to the image signal in the photon-counting system), by assuming a Poisson distribution, and error propagation through the interpolation process. In summary,

\[
\sigma_{\text{intra}}^2 = \sigma_{\text{ROI}}^2 + \sigma_{\text{quant}}^2 \quad \text{and} \quad \sigma_{\text{inter}}^2 = \sigma_{\text{drift}}^2 + \sigma_{\text{quant}}^2.
\] (4)

Data from all ROI locations and images (four-by-four individual readings) were added in order to estimate the expectation value for each sample. The spread between the expectation values for different samples is referred to as the total variability (\(\sigma_{\text{tot}}\)), which includes variation between samples (\(\sigma_{\text{inter}}\)) fluctuations between measurements (\(\sigma_{\text{drift}}\)), random measurement errors from thickness uncertainties as described in section 2.4 (\(\sigma_{\text{th}}\)), and the total quantum noise for the sample (\(\sigma_{\text{quant}}\)):
\[
\sigma_{\text{tot}}^2 = \sigma_{\text{inter}}^2 S + \frac{1}{16} \times \sigma_{\text{drift}}^2 + \sigma_{\text{th}}^2 + \sigma_{\text{quant}}^2 S, \quad \text{where} \quad \sigma_{\text{quant}}^2 = \frac{1}{16} \times \sigma_{\text{quant}}^2 I.
\]

(5)

The intra-image variability is not included in the total variability because the ROIs generally do not cover the entire lesion; \(\sigma_{\text{ROI}}\) is therefore not a complete measure of sample inhomogeneity but rather a measure of the quality of the ROI selection. Sample inhomogeneity is instead captured by \(\sigma_{\text{inter}}\).

Two-sample \(t\)-tests and chi-square variance tests were used to test the differences between the means and the variances of the different measurements.

3. Results

Of the original 84 samples, 23 were excluded from the study: 6 cases were DCIS with no mass lesion discernible on the images, and 17 cases were excluded for technical reasons, mainly because the pathology lab detected that tumour was not histologically present at both cut surfaces despite initial appearances of the fixed specimen block. Of the remaining 61 biopsy proven solid fixed tissue specimens, 37 were invasive ductal carcinoma (3 grade I, 19 grade II, 15 grade III), 14 lobular carcinoma, 5 special type (2 papillary carcinoma, 2 mucinous, and 1 metaplastic), and 5 benign (4 fibroadenomas and 1 phyllodes tumour). Due to the logistical constraints of obtaining tissue from, and returning it to, the pathology laboratory cut up room, without disruption or delaying processing, final sample eligibility could only be verified after imaging had been performed, leading to the relatively large number of exclusions.

The mean sample thickness was 6.4 mm, and the mean total sample ROI size (sum of four sub ROIs) was 54 mm². The systematic error caused by uncertainties in thickness and density of the measurement setup was estimated according to section 2.4 for an average sample to be 0.7% in the Al- and PMMA-equivalent thicknesses. The random error caused by variations in thickness over the measurement area and variation in the sample thickness measurements (\(\sigma_{\text{th}}\)) was estimated for an average sample to be 0.9% and 0.5% in the Al- and PMMA-equivalent thicknesses, respectively.

Figure 3 shows measurement results for the 61 solid (5 benign, 56 malignant) samples that were included in this study, as well as for 50 cyst-fluid samples and 50 water samples from Fredenberg et al (2013), all expressed in terms of the equivalent Al and PMMA thicknesses and normalized to a sample thickness of 10 mm and a PMMA density of 1.19 g cm⁻³. The aspect ratio of the \(x\) and \(y\) axes in figure 3 (1:10) corresponds approximately to the relative attenuation length of Al and PMMA at mammography energies.

The left panel of figure 3 shows an overview of the Al-PMMA vectors with the total variability (\(\sigma_{\text{tot}}\), one standard deviation) indicated as error bars at the end of each vector. The angle (\(\theta\)) and magnitude (\(r\)) are illustrated on the cyst-fluid vector. Note that the Al and PMMA axes in figure 3 are transposed compared to some previous studies (Johns and Yaffe 1985, 1987), but we have kept the definition of \(\theta\) (denoted \(\Phi\) by Johns and Yaffe) to facilitate comparison, which is why the angle runs from the ordinate rather than from the abscissa in figure 3. The (effective) atomic number of the sample material determines \(\theta\), and the angles for \(Z = 7\) and \(Z = 8\) are indicated in figure 3 for illustration.

The right panel of figure 3 shows a close-up of the measurements. For the solid lesions, individual measurement points are shown shaded in grey with error bars for the inter-image variability (\(\sigma_{\text{inter}}\)). The 5 benign lesions are circled. For the previously published cyst fluid and water measurements, the perimeters (convex hulls) of the set of individual measurement points are shown. The mean values and total variabilities (\(\sigma_{\text{tot}}\)) of each sample type are shown.
as bold error bars. The smallest $\theta$ of the cyst fluid samples is indicated with a red dotted line and the most extreme outlier of the solid lesions toward the cyst fluid distribution is indicated with an arrow. All mean values and variability measures are listed in table 1.

In common with Tomal et al (2010) we have bundled the data for different types of malignant lesions. There was no significant difference in attenuation between benign and malignant solid breast lesions ($P > 0.5$ determined by two-sample $t$-tests on the PMMA- and Al-equivalent thicknesses), which is also manifested by the almost complete overlap by the mean values for benign solids and malignant solid lesions in figure 3.

Solid breast lesion attenuation (benign and malignant) was significantly different from cyst-fluid attenuation and significantly different from water attenuation in terms of PMMA- and Al-equivalent thicknesses ($P < 0.001$, determined by two-sample $t$-tests). Solid breast lesion attenuation was significantly different from cyst attenuation in terms of angle ($\theta$) and magnitude ($r$) of the Al-PMMA vector and significantly different from water attenuation in terms of $r$ ($P < 0.001$, determined by two-sample $t$-tests), but there was no significant difference from water attenuation in terms of $\theta$ ($P > 0.5$, determined by a two-sample $t$-test). The small difference between water and solid lesions in terms of $\theta$ is manifested by the Al-PMMA vector for solid lesions in figure 3 being obscured by the water vector for most but not all of its length as the $r$ values are different.

The random fluctuations between measurements ($\sigma_{\text{inter}}$) and the sample variability ($\sigma_{\text{intra}}$) were not significantly different from the expected quantum noise between ROIs ($P > 0.1$, determined by two-sample $t$-tests between $\sigma_{\text{inter}}$/$\sigma_{\text{intra}}$ and $\sigma_{\text{quant}}$). The total variability of the
solid samples ($\sigma_{\text{tot}}$) was, however, significantly larger than the expected quantum noise, $\sigma_{\text{quant \ S}}$ ($P < 0.001$, determined by chi-square variance tests on the PMMA- and Al-equivalent thicknesses). The total variability of the solid malignant samples was 3 (Al) and 4 (PMMA) times larger than that of the cyst fluid samples, and 16 (Al) and 7 (PMMA) times larger than for the water samples.

The linear attenuation coefficients of solid breast lesions, calculated from the measured PMMA- and Al-equivalent thicknesses at a range of x-ray energies relevant to mammography, are shown in table 2 and plotted in figure 4. (Note that figure 4 does not include the benign lesions as the difference to malignant lesions was too small to be visualized in the plot.)

Previously published linear attenuation coefficients of cyst fluid and water, calculated in an equivalent manner (Fredenberg et al 2013), are included in table 2 and figure 4 for reference.
Figure 4 also plots the linear attenuation coefficients of solid lesions and cyst fluid normalized to the linear attenuation coefficient of water in order to better visualize the difference in attenuation between the materials.

4. Discussion

4.1. Measurement results

4.1.1. Differentiation between tissues with spectral imaging. The challenge of distinguishing between different material types in regular x-ray imaging is that it is not possible to differentiate between a thin piece of highly attenuating material and a correspondingly thicker piece of lower attenuating material. Spectral imaging circumvents this problem by considering not only the actual attenuation, but also the energy dependence of the attenuation. It may therefore be possible to distinguish between cyst fluid and solid lesions despite the close-to overlapping linear attenuation curves in the upper panel of figure 4 (less than 2% difference on average in the 15–40 keV energy interval) because the shapes of the curves are different (manifested as different slopes in the lower panel of figure 4). Water and solid lesions, on the other hand, may be much more difficult to differentiate between even in spectral imaging, despite the 7% difference in attenuation, because the energy dependence of the linear attenuation is close-to identical (parallel flat curves for solid and water attenuation in the lower panel of figure 4).

The differences of water and cyst fluid attenuation relative to that of solid lesions are further illustrated by the magnitude ($r$) and angle ($\theta$) of the Al-PMMA vector. As defined toward the end of section 2.2, $r$ is directly proportional to the thickness of the sample and therefore cannot be measured unless the thickness of the sample is known. Such an independent thickness measurement was used in the present pre-clinical study, but may not be available in clinical applications of spectral imaging. Contrary to $r$, $\theta$ is independent of the sample thickness and can be measured with spectral imaging (but not with conventional x-ray imaging) without any a priori information. It was found in this study that $r$ of water and cyst fluid are both significantly lower than those of solid lesions, whereas $\theta$ is equal for water and solid lesions but significantly higher for cyst fluid compared to solid lesions. It appears that solid lesions have an effective atomic number similar to that of water, but higher density. The effective atomic number of cyst fluid seems slightly higher than that of solid lesions and water.

| Table 2. Linear attenuation coefficients of solid breast lesions (benign and malignant) calculated from the measured PMMA- and Al-equivalent thicknesses (bold text), and compared to previously published data on cyst fluid and water. |
| --- |
| Photon energy (keV): | 15 | 20 | 25 | 30 | 35 | 40 |
| Malignant solid linear attenuation (cm⁻¹): | 1.76 | 0.859 | 0.541 | 0.400 | 0.327 | 0.286 |
| Benign solid linear attenuation (cm⁻¹): | 1.78 | 0.864 | 0.544 | 0.402 | 0.328 | 0.286 |
| Cyst fluid linear attenuation (cm⁻¹): | 1.76 | 0.852 | 0.533 | 0.391 | 0.318 | 0.277 |
| Water linear attenuation (cm⁻¹): | 1.64 | 0.800 | 0.504 | 0.372 | 0.305 | 0.266 |

Note: The total variability (one standard deviation) is given in parenthesis.
Despite the large spread, no solid samples fall within the shaded region of the cyst distribution (figure 3). Nevertheless, 16 malignant (29% of the total number) and 2 benign (40% of the total number) samples overlap with the cyst distribution in terms of $\theta$ (figure 3, area below the dotted line), and may therefore be challenging to distinguish from cyst fluid without providing thickness information. One (1) solid sample (figure 3, arrow) falls ‘on the other side’ of the cyst distribution with a higher $\theta$ than any of the cyst fluid cases.

A challenge for distinguishing between different material types in clinical applications of spectral imaging is the influence of overlapping tissue that obscures the lesion. We have developed a method that circumvents that problem (Norell et al 2012), and a clinical study based on the technique has been initiated at the Cambridge Breast Unit (Erhard et al 2015). In short, upon detection of a suspicious lesion, the local breast composition (the amounts of adipose and fibro-glandular tissue) is assessed in a reference region surrounding the lesion using spectral information and knowledge of the attenuation of normal breast tissue. By assuming these two properties to be constant or varying in a predictable way over the lesion, the influence of superimposed tissue can be removed. Subsequently, with knowledge of the attenuation of cysts and tumors, the lesion thickness and contents (cyst or tumor) may be determined.

4.1.2. Total variability. Compared to the previous measurements on cyst fluid, the spread between the different samples of solid breast lesions was large. The random measurement errors (section 2.4) were substantially larger for the measurement of solid lesion attenuation in this study compared to the previous measurements on cyst fluid and water, which is mainly due to the repeated thickness measurements and to the thinner samples. The random
fluctuations within each ROI ($\sigma_{\text{intra I}}$) and between repetitive measurements ($\sigma_{\text{inter I}}$) were not significantly different from the expected quantum noise ($\sigma_{\text{quant I}}$), which indicates that quantum noise was the dominating noise source within a measurement, and that the sample homogeneity in each ROI and the system stability were good, i.e. $\sigma_{\text{ROI}} \ll \sigma_{\text{quant I}}$ and $\sigma_{\text{drift}} \ll \sigma_{\text{quant I}}$. The expected quantum noise was almost a factor of two higher for the solid lesions compared to the cyst fluid measurements, again because of the thinner samples.

For the total variability ($\sigma_{\text{tot}}$), the random thickness errors of the measurement setup ($\sigma_{\text{th}}$) and quantum noise ($\sigma_{\text{quant/uni00A0S}}$) together constitute approximately 30% of the variability in PMMA- and 20% of the variability in Al-equivalent thicknesses. In contrast, a similar estimation for the water measurements, for which sample variations can be expected to be at a minimum, yields contributions by random measurement errors and quantum noise of more than 90%. It is reasonable to expect that some part of the variability comes from contamination of other tissue types in the samples (mainly adipose and fibro-glandular tissue). Care was, however, taken to minimize this effect (samples without clear tumour surfaces on both sides were excluded, and the ROIs were selected in homogeneous areas of the sample) and sample homogeneity within each ROI was found to be good. We therefore expect the major part of the total variability to be caused by the natural variation of tumour tissue.

In common with previous studies, we used PMMA and Al as basis materials, but it is conceivable to use any other pair of (low-atomic-number) materials as basis set without affecting any of the basic results, including the discrimination between different tissue types or the calculation of linear attenuation. One reason for going to another set of basis materials would be to span a larger range of attenuation and avoiding negative thicknesses; Al and polyethylene would, for instance, cover the entire range of normal breast tissue (adipose and glandular) and breast lesions, i.e. tumour tissue and cyst fluid (Erhard et al 2015). Another reason for choosing different basis materials would be if any of the materials were limiting the accuracy or precision of the measurement, for instance, because of machining defects. In our case, the random thickness error ($\sigma_{\text{th}}$) was estimated to be larger for Al (0.9% compared to 0.5% for PMMA), and it is possible that some precision could be gained by substituting Al for a different basis material such as a plastic.

4.2. Comparison to other studies

4.2.1. Malignant lesions. Figure 5 shows a comparison between the data presented in this study and most of the attenuation data on breast tumors available in the literature (Johns and Yaffe 1987, Chen et al 2010, Tomal et al 2010). The data from Carroll et al (1994) were not tabulated in the paper and are therefore not included in figure 5. The equivalent Al and PMMA thicknesses for the published data were found by fitting to the linear attenuation coefficients according to equation (3). For all three attenuation data sets, the average residuals of the fitted curve compared to the original one were within 0.005% in the considered energy range. The values calculated from each published study are shown as dark blue open markers, and the mean and standard deviation of all previously published studies are shown as a dark blue error bar. The data from the present study are shown as the convex hulls, and solid and dotted error bars that indicate the mean values and the total variabilities of the malignant and benign lesion distributions. The mean and standard deviation of all published studies, including the present study, are shown as a bold green error bar. The interpretation of the cyan markers in figure 5 is explained in the following.

Johns and Yaffe (1987) published the equivalent Al and PMMA thicknesses fitted over the range 18 to 110 keV and these data (renormalized to a thickness of 10 mm and Al and
PMMA densities of 2.699 g/cm$^3$ and 1.19 g/cm$^3$ are included in figure 5 for comparison (cyan diamond). There are clear differences in mean values compared to our fit in the mammography energy range (dark blue diamond). Equation (1) is an approximation and results differ somewhat when fitting over different energy ranges, but that cannot explain the discrepancy; recalculating the linear attenuation from the fit yields an average residual of $-0.7\%$ compared to the published linear attenuation over the entire energy interval, whereas a recalculation from the published Al and PMMA thicknesses yields an average residual of 4.0%. This is substantially larger than the maximum fitting error reported by Johns and Yaffe (0.2%), and we therefore expect the major part of the discrepancy to come from errors in the linear attenuation values of Al and PMMA. Firstly, the coherent and incoherent scattering cross sections for compounds are not exactly the weighted sums of the cross sections for individual elements, which affects calculation of the PMMA attenuation. Further, it is conceivable that systematic differences between elemental cross section tabulations have contributed to the error; Johns and Yaffe used the tabulations by Plechaty et al (1975) whereas we used those of the XCOM database (Berger et al 2010). Errors in elemental cross section tabulations are typically a few percent and hence not negligible (Saloman et al 1988, Hubbell 1999).

The equivalent material thicknesses for malignant solid lesions presented in the present study (dark grey convex hull and black solid error bar) are comparable to, and reasonably close to, the mean of previously published data (dark blue markers and error bar). The range of the previously published data is, however, relatively wide. One part of the spread may be caused by randomness within each study, including random measurement errors, quantum noise, and natural variation of tumor tissue. Johns and Yaffe (1987) published the range (maximum and minimum values) of their measured Al and PMMA thicknesses, which are plotted in figure 5 (cyan diamond and triangles) and gives some indication of the contribution from
random errors. Our data on malignant solid lesions cover a wider range (dark grey convex hull), but the set is also substantially larger, which generally increases the range because of outliers but does not affect the range in terms of the standard deviation. (Note that the range of the smaller set of benign lesions from this study—light grey convex hull in figure 5—is similar to that of Johns and Yaffe, but the standard deviation for the benign lesions—dotted error bar—is comparable to our results for malignant lesions—solid black error bar.) Chen et al (2010) and Tomal et al (2010) reported values of the standard deviation for their measured linear attenuation coefficients. Averaging that data in the range 15 to 30 keV yields values of 0.019 cm\(^{-1}\) (Chen et al) and 0.061 cm\(^{-1}\) (Tomal et al), which may be compared to our data on the standard deviation in table 2 and an average value of 0.022 cm\(^{-1}\) for the same energy range. We therefore expect the spread between measurements (the total variation) seen in the present study to be of the same order as that of previous studies.

If random fluctuations within each published study would be the sole contributor to the spread between the mean values, and assuming equal random errors in the present study and in the published studies on average, the spread (standard deviation) between the published mean values would be smaller than the total variation reported in this study because the mean values are better estimates of the expectation values than are the individual measurements. The spread between published mean values is, however, approximately a factor of two larger than the total variation of the present study, and it is likely that a substantial part is caused by systematic differences between the measurement setups. We estimated our systematic measurement errors to be 0.7%, and it is likely that other studies end up at similar values, but there may also be other systematic differences between the studies, including differences in tissue handling and tumor types, and in our case, the aforementioned uncertainty in the elemental cross section tabulations. One way of circumventing these types of systematic uncertainties for quantitative analyses is to perform all measurements within the same framework. We have therefore measured tumor attenuation with the same method as previously applied to cyst fluid and under conditions that are as similar as possible to the screening environment where potential future applications will be used.

In summary, the spread in the available attenuation data may be caused by (1) a large natural spread between samples, (2) random measurement errors, and (3) different experimental conditions in the different studies that cause systematic differences and errors. Uncertainty number (1) and (2) may be reduced by adding samples to the available data. The present study contributes a relatively large sample set (56 samples) compared to published studies (~38 samples in total) and may help in that respect. Further, assuming that the systematic errors within each study are random between the studies, simply adding more studies to the literature will help reduce uncertainty number 3. Including the present study shifts the average of published data slightly (green error bar in figure 5), and it is fair to assume that to be a better estimate of the true expectation value.

4.2.2. Benign lesions. Tomal et al (2010) measured the attenuation of 6 fibroadenomas, and the calculated equivalent Al and PMMA thicknesses are shown in figure 5 (cyan square and cross). There is a clear difference to the mean value of benign lesions reported in this study (dotted error bar), which is similar to the difference between malignant samples in the two studies (solid black error bar and dark blue square) and is likely caused by systematic differences between the measurements. Further, the data reported by Tomal et al suggest slightly lower Al and PMMA thicknesses compared to malignant samples, as opposed to our study, where the Al and PMMA thicknesses were rather higher or equal to the malignant samples. Similar to our study, however, Tomal et al reported that the difference was not significant, and these discrepancies may be caused by statistical errors. It should be noted that three samples...
of benign lesions were included also in the study by Johns and Yaffe (1987), but data for these samples were not tabulated separately.

4.2.3. Tissue fixation. In this study, formalin fixed tissue was used as opposed to fresh tissue, which is a potential bias when making direct interpretation of the results to clinical mammography. Chen et al (2010) measured the linear attenuation coefficients in the energy range 17 to 23 keV of 6 of their malignant samples before and after formalin fixation. The resulting fitted Al- and PMMA-equivalent thicknesses are shown in figure 5 (cyan circles). The mean value of the fixed tissue (cyan open circle) is clearly different from that of the full study (dark blue circle). This difference may be caused by the use of a smaller number of samples and the narrow energy span. More interesting is the difference between fixed (cyan open circle) and fresh (cyan circle and cross) tissue within the subset; fresh tissue exhibits a higher Al thickness and a lower PMMA thickness than fixed tissue, which moves the solid lesions closer to the attenuation of cyst fluid. This is slightly worrying for the application of separating solid lesions from cysts, but it is currently not clear how significant the difference between fresh and fixed tissue is considering in particular the calculation of Al and PMMA thicknesses within the limited energy range.

5. Conclusions and outlook

A significant separation between cyst fluid and tumour attenuation was found on average, which suggests it may be possible to distinguish cystic from solid breast lesions using screening with spectral mammography, raising the exciting possibility of reducing unnecessary recalls. As our study measured tumour attenuation under conditions that were as similar as possible to the screening environment and with the same spectral method as a previous study on cyst-fluid attenuation, we expect the comparison between cysts and tumours to be valid and applicable to screening applications. These results therefore lay the groundwork for a clinical trial, which is currently under way.

Previously published data on the x-ray attenuation of solid breast tumours cover a relatively wide range, which is likely to be caused by (1) a large natural spread between samples, (2) random measurement errors, and (3) different experimental conditions in the different studies that cause systematic differences and errors. The present study adds a relatively large sample set to the published data and may contribute to reduce the overall uncertainty in the literature.

There was a relatively large spread between the different samples of solid breast lesions. We expect the major part of the spread to be caused by natural variation between samples and only to a minor extent by random measurement errors. No significant difference in attenuation was found between benign and malignant solid lesions.

Our future work will focus on a clinical trial to further investigate the possibility of distinguishing cysts from tumours in the screening image. In addition, our pre-clinical study will continue in order to clarify the difference in x-ray attenuation between fixed and fresh tissue, and to measure the attenuation of healthy adipose and fibro-glandular breast tissue, which is also a necessary prerequisite for distinguishing cysts from tumours in the clinical setting where the lesions are obscured by normal tissue.

Acknowledgments

Special thanks are extended to Dr E Provenzano, Breast Histopathologist, and Mr James Neal, Advanced Practitioner in Breast Dissection, both at the Department of Histopathology.
Addenbrooke’s Hospital and Cambridge NIHR Biomedical Research Centre, for their support in preparing the tissue samples, and to Dr Miriam von Tiedemann, Philips Health Systems, for coordinating parts of the study and for help in methodology development. The authors would also like to thank Dr Björn Cederström and Dr Klaus Erhard, both with Philips Health Systems, and Mr Staffan Karlsson at the Royal Institute of Technology (KTH) for constructing the sample holder. Part of this work was carried out within the OPTIMAM2 project funded by Cancer Research UK (grant number C30682/A17321). The Cambridge Human Research Tissue Bank is supported by the NIHR Cambridge Biomedical Research Centre.

References

Alvarez R E and Macovski A 1976 Energy-selective reconstructions in x-ray computerized tomography Phys. Med. Biol. 21 733–44
Åslund M, Cederström B, Lundqvist M and Danielsson M 2006 Scatter rejection in multi-slit digital mammography Med. Phys. 33 933–40
Berger M J, Hubbell J H, Seltzer S M, Chang J, Coursey J S, Sukumar R, Zucker D S and Olsen K 2010 XCOM: Photon Cross Section Database (Gaithersburg, MD: National Institute of Standards and Technology) (http://physics.nist.gov/xcom)
Berglund J, Johansson H, Lundqvist M, Cederström B and Fredenberg E 2014 Energy weighting improves dose efficiency in clinical practice: implementation on a spectral photon-counting mammography system J. Med. Imaging 1 31003
Boone J M, Fewell T R and Jennings R J 1997 Molybdenum, rhodium, and tungsten anode spectral models using interpolating polynomials with application to mammography Med. Phys. 24 1863–74
Brett J and Austoker J 2001 Women who are recalled for further investigation for breast screening: psychological consequences 3 years after recall and factors affecting re-attendance J. Public Health Med. 23 292–300
Brewer N T, Salz T and Lille S E 2007 Systematic review: the long-term effects of false-positive mammograms Ann. Intern. Med. 146 502–10
Carroll F E et al 1994 Attenuation of monochromatic x-rays by normal and abnormal breast tissues Investigative Radiol. 29 266–72
Chen R C et al 2010 Measurement of the linear attenuation coefficients of breast tissues by synchrotron radiation computed tomography Phys. Med. Biol. 55 4993–5005
Ding H and Molloi S 2012 Quantification of breast density with spectral mammography based on a scanned multi-slit photon-counting detector: a feasibility study Phys. Med. Biol. 57 4719–38
Erhard K, Kilburn-Toppin F, Willsher P, Moa E, Fredenberg E, Wiberneit N, Buelow T and Wallis M G 2015 Characterization of cystic lesions by spectral mammography: results of a clinical pilot study Investigative Radiol. (doi: 10.1097/RLI.0000000000000246)
Fredenberg E, Åslund M, Cederström B, Lundqvist M and Danielsson M 2010b Observer model optimization of a spectral mammography system Proc. SPIE Medical Imaging 2010: Physics of Medical Imaging (San Diego) ed E Samei and N J Pelc vol 7622 p 762210
Fredenberg E, Dance D R, Willsher P, Moa E, von Tiedemann M, Young K C and Wallis M G 2013 Measurement of breast-tissue x-ray attenuation by spectral mammography: first results on cyst fluid Phys. Med. Biol. 58 8609–20
Fredenberg E, Lundqvist M, Cederström B, Åslund M and Danielsson M 2010a Energy resolution of a photon-counting silicon strip detector Nucl. Instrum. Methods A 613 156–62
Guerriero C, Gillan M G C, Cairns J, Wallis M G and Gilbert F J 2011 Is computer aided detection (CAD) cost effective in screening mammography? A model based on the CADET II study BMC Health Serv. Res. 17 11
Hubbell J H 1999 Review of photon interaction cross section data in the medical and biological context Phys. Med. Biol. 44 R1–22
Johns P C and Yaffe M J 1985 Theoretical optimization of dual-energy x-ray imaging with application to mammography Med. Phys. 12 289–96
Johns P C and Yaffe M J 1987 X-ray characterisation of normal and neoplastic breast tissues Phys. Med. Biol. 32 675–95
Kappadath S C and Shaw C C 2008 Quantitative evaluation of dual-energy digital mammography for calcification imaging Phys. Med. Biol. 53 5421–43
Lehmann L A, Alvarez R E, Macovski A, Brody W R, Pelc N J, Riederer S J and Hall A L 1981 Generalized image combinations in dual KVP digital radiography Med. Phys. 8 659–67
Maxwell A J, Beattie C, Lavelle J, Lyburn I, Sinnatamby R, Garnett S and Herbert A 2013 The effect of false positive breast screening examinations on subsequent attendance: retrospective cohort study J. Med. Screening 20 91–8
Norell B, Fredenberg E, Leifland K, Lundqvist M and Cederström B 2012 Lesion characterization using spectral mammography Proc. SPIE Medical Imaging 2012: Physics of Medical Imaging (San Diego) ed N J Pelc et al vol 8313 p 83130I
Plechaty E F, Cullen D E and Howerton R J 1975 Tables and Graphs of Photon Interaction Cross Sections from 1.0 keV to 100 MeV Derived from the LLL Evaluated Nuclear Data Library vol 6 (Livermore, CA: Lawrence Livermore Laboratory)
Román R et al 2011 Effect of false-positives and women’s characteristics on long-term adherence to breast cancer screening Breast Cancer Res. Treat. 130 543–52
Saloman E B, Hubbell J H and Scofield J H 1988 X-ray attenuation cross sections for energies 100 eV to 100 keV and elements $Z = 1$ to $Z = 92$ At. Data Nucl. Data Tables 38 1–196
Smith-Bindman R et al 2003 Comparison of screening mammography in the United States and the United Kingdom JAMA 290 2129–37
Taibi A, Fabbri S, Baldelli P, Di Maggio C, Gennaro G, Marziani M, Tuffanelli A and Gambaccini M 2003 Dual-energy imaging in full-field digital mammography: a phantom study Phys. Med. Biol. 48 1945–56
Tomal A, Mazarro I, Kakuno E M and Poletti M E 2010 Experimental determination of linear attenuation coefficient of normal, benign and malignant breast tissues Radiat. Meas. 45 1055–9