Ultrasound in Med. & Biol., Vol. ■, No. ■, pp. 1–9, 2016

0301-5629/$ - see front matter

ELSEVIER

http://dx.doi.org/10.1016/j.ultrasmedbio.2016.09.007

● *Original Contribution*

METROLOGICAL VALIDATION OF A MEASUREMENT PROCEDURE FOR THE CHARACTERIZATION OF A BIOLOGICAL ULTRASOUND TISSUE-MIMICKING MATERIAL

Taynara Q. Santos, André V. Alvarenga, Débora P. Oliveira, and Rodrigo P. B. Costa-Felix
Laboratory of Ultrasound, Directory of Scientific and Industrial Metrology, National Institute of Metrology, Quality and Technology, Rio de Janeiro, Brazil

(*Received* 6 *July* 2016; *revised* 6 *September* 2016; *in final form* 6 *September* 2016)

Abstract—The speed of sound and attenuation are important properties for characterizing reference materials such as biological phantoms used in ultrasound applications. There are many publications on the manufacture of ultrasonic phantoms and the characterization of their properties. However, few studies have applied the principles of metrology, such as the expression of the uncertainty of measurement. The objective of this study is to validate a method for characterizing the speed of sound and the attenuation coefficient of tissue-mimicking material (TMM) based on the expression of the measurement of uncertainty. Six 60-mm-diameter TMMs were fabricated, three 10 mm thick and three 20 mm thick. The experimental setup comprised two ultrasonic transducers, acting as transmitter or receiver depending on the stage of the measurement protocol, both with a nominal center frequency of 5 MHz and an element diameter of 12.7 mm. A sine burst of 20 cycles and 20-V peak-to-peak amplitude at 5 MHz excited the transmitter transducer, producing a maximum pressure of 0.06 MPa. The measurement method was based on the through-transmission substitution immersion technique. The speed of sound measurement system was validated using a calibrated stainless-steel cylinder as reference material, and normalized errors were <0.8. The attenuation coefficient measurement method was validated using replicated measurements under repeatability conditions. The normalized error between the two measurement sets was <1. The proposed uncertainty models for the measurements of the speed of sound and the attenuation coefficient can help other laboratories develop their own uncertainty models. These validated measurement methods can be used to certify a TMM as a reference material for biotechnological applications. (E-mail: avalvarenga@inmetro.gov.br) © 2016 World Federation for Ultrasound in Medicine & Biology.

Key Words: **Ultrasound, Tissue-mimicking material, Speed of sound, Attenuation coefficient, Validation, Metrology, Measurement uncertainty.**

INTRODUCTION

According to the International Organization for Standardization Committee on Conformity Assessment (ISO/IEC 2005), the validation of a measurement procedure is defined as "the confirmation by examination and provision of objective evidence that the particular requirements for a specific intended use are fulfilled." Therefore, validation ensures that the measurement procedure is adequate for its intended use (BIPM 2012). There are many published measurement methods for characterizing the ultrasonic or acoustic properties of materials (Zeqiri et al. 2010). In particular, there have been many studies on the ultrasonic properties of phantoms, such as the speed of sound and the attenuation coefficient, with respect to their manufacture and characterization (Bader et al. 2016; Brewin et al. 2008; Browne et al. 2003; Inglis et al. 2006; Madsen et al. 1978, 1998; Rajagopal et al. 2015; Sun et al. 2012; Teirlinck et al. 1998). However, few studies have applied the principles of metrology, such as the expression of the uncertainty of measurement and the use of validated measurement methods. Therefore, it is important that laboratories have the means and objective criteria to show that the procedures used for measuring ultrasonic properties such as the speed of sound and the attenuation coefficient are valid, and that the tests performed lead to reliable and appropriate results (ISO/IEC 2010).

Rajagopal et al. (2015) reported that the speed of sound in a 3.03-mm-thick tissue-mimicking material

---

Address correspondence to: André V. Alvarenga, Laboratory of Ultrasound, Division of Acoustic and Vibration Metrology, Directory of Scientific and Industrial Metrology, Av. N. Sra. das Graças, 50 – Xerém, Duque de Caxias, Rio de Janeiro 25250-020, Brazil. E-mail: avalvarenga@inmetro.gov.br

(TMM) was 1548 m/s with an uncertainty of 6.1 m/s. Brewin et al. (2008) conducted a 2-y stability study of the acoustic properties of an agar-based TMM using two measuring systems: one used narrowband transmission reception and the other used broadband pulse-echo. Both systems were based on the replacement technique with a center frequency of 20 MHz. Using a pre-clinical ultrasound scanner and a scanning acoustic macroscope (SAM), Sun et al. (2012) characterized the ultrasonic properties of the agar-based TMM in the 10–47 MHz range by measurements based on a broadband reflection substitution technique. To validate the SAM for measuring the speed of sound, Cannon et al. (2011) used a silicone oil reference cell (provided by the National Physical Laboratory, Middlesex, UK) with the pulse-echo method at 7.5 MHz in a tank containing degassed water at 21.5°C. Browne et al. (2003) developed a method to characterize the attenuation coefficient of four different types of TMM, including that described in IEC 60601 (2015), in the 2.25–15 MHz frequency range. Inglis et al. (2006) designed and constructed an anthropomorphic model esophagus suitable for use with endoscopic ultrasound. Different agar-based TMMs were fabricated to reproduce the ultrasonic properties of different anatomic and pathologic structures. The ultrasonic properties were measured using a SAM at frequencies between 7.5 MHz and 12 MHz.

This paper reports the metrological validation of the measurement procedure, developed at the Laboratory of Ultrasound of the Institute of Metrology, Quality, and Technology, to characterize the speed of sound and the attenuation coefficient of biotechnological materials. Validation is based on the expression of the uncertainty of measurement as proposed by the Guide to the Expression of Uncertainty in Measurement (BIPM 2008). The reference testing body was a standardized TMM (IEC 2015), typically used in safety testing and the calibration of ultrasound equipment for clinical use.

MATERIAL AND METHODS

Experimental model

Six 60-mm-diameter TMMs were fabricated according to IEC 60601-2-37 (IEC 2015), three 10 mm thick and three 20 mm thick. The materials and their appropriate mass percentage used to fabricate the TMM phantoms are specified in Table 1 (IEC 2015; Teirlinck et al. 1998). Glycerol provides the TMM with the required speed of sound, benzalkonium chloride prevents the growth of micro-organisms, 0.3-μm alumina ($Al_2 O_3$) affects primarily the TMM attenuation coefficient, while silicon carbide and 3-μm Al_2O_3 produce backscatter to mimic biological tissue (Ramnarine et al. 2001). Details about the TMM fabrication process are provided in Teirlinck et al. (1998) and IEC (2015), and the specific procedure followed for fabricating the TMM in this work is described in Souza et al. (2016).

Table 1. Materials and amount used for the TMM described in Teirlinck et al. (1998) and IEC (2015)

Material	Mass [%]
Alumina (Al_2O_3) (0.3 μm)	0.88
Alumina (Al_2O_3) (3 μm)	0.94
Agar	3.02
Glycerin	11.21
H_2O	82.95
Benzalkonium chloride	0.47
Silicon carbide	0.53
Total	100.00

TMM = tissue-mimicking material.

The six TMMs were randomly grouped into three sets, with each pair containing one 20-mm-thick and one 10-mm-thick TMM. Each pair underwent five measurements under repeatability conditions to characterize the ultrasonic longitudinal speed of sound and the attenuation coefficient. The experimental procedure was validated under repeatability and intermediate precision conditions. As indicated in the standards, intermediate precision conditions of measurement can be achieved with the use of two operators and an interval between subsequent measurements (BIPM 2012; ISO 2001). Thus, in this study, two different operators repeated the same set of measurements on the same samples 15 d after the initial set of measurements and under the same experimental conditions.

Speed of sound

The experimental setup used to measure the speed of sound consisted of two single-crystal circular immersible piezoelectric unfocused ultrasonic transducers (T_{x1} and T_{x2}) that acted as transmitter or receiver at different stages of the measurement protocol; both had a nominal center frequency of 5 MHz and a 12.7-mm-diameter element (V303, Panametrics-NDT, Olympus, Waltham, MA, USA). Both transducers were previously scanned and their beam width at focal point was about 3.5 mm (standard uncertainty of 0.1 mm), while at 3 dB after the focal point, the beam width was about 6.5 mm (standard uncertainty of 0.1 mm) (Alvarenga et al. 2016).

The transducers and the specimen were mounted on positioning systems with 5 degrees of freedom to ensure adequate alignment. The transducers and TMMs were placed in a tank of deionized water for 30 min to attain thermal equilibrium. An arbitrary waveform generator (33250 A, Agilent Technologies, Santa Clara, CA, USA) excited the emitter transducer with a sine burst (Brewin et al. 2008) of 20 cycles and a 20-V peak-to-peak amplitude at 5 MHz, producing a maximum