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● *Review*

NEONATAL CRANIAL ULTRASOUND: ARE CURRENT SAFETY GUIDELINES APPROPRIATE?

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Abstract—Ultrasound can lead to thermal and mechanical effects in interrogated tissues. We reviewed the literature to explore the evidence on ultrasound heating on fetal and neonatal neural tissue. The results of animal studies have suggested that ultrasound exposure of the fetal or neonatal brain may lead to a significant temperature elevation at the bone–brain interface above current recommended safety thresholds. Temperature increases between 4.3 and 5.6°C have been recorded. Such temperature elevations can potentially affect neuronal structure and function and may also affect behavioral and cognitive function, such as memory and learning. However, the majority of these studies were carried out more than 25 y ago using non-diagnostic equipment with power outputs much lower than those of modern machines. New studies to address the safety issues of cranial ultrasound are imperative to provide current clinical guidelines and safety recommendations. (E-mail: Michal.schneider@monash.edu) **© 2016 World Federation for Ultrasound in Medicine & Biology.**

Key Words: **Cranial ultrasound, Fetal brain heating, Preterm infants, Thermal index, Mechanical index, Ultrasound safety.**

INTRODUCTION

Neonatal cranial ultrasound is the mainstay of modern management for prematurely delivered babies and those born after a complicated delivery involving episodes of hypoxia. The number of preterm births worldwide has steadily increased in the last 10 y, accounting for 11.1% of all live births (Blencowe et al. 2012). The developing brain of premature infants is vulnerable to injury in the early postnatal period (Ballabh 2010; Inder and Volpe 2000; Rorke 1992; Volpe et al. 2011). Cranial ultrasound is used routinely to monitor the development and complications of hemorrhagic and ischemic brain injury in these newborns in the weeks after delivery.

Despite the absence of ionizing radiation, ultrasound has the potential to cause focally induced temperature increases in the tissues being interrogated. Although several studies on this topic have emerged, only one review has been published to date specifically reporting on the implications of ultrasound-induced temperature increases on the developing neonatal brain in the human. To this end, we identified all relevant empirical studies on the physical interaction of ultrasound with developing neural tissue both *in utero* and during the postnatal period.

The focus of this review is on the relevance of the available evidence to the human neonatal brain and, importantly, on the clinical relevance of these studies for modern ultrasound machines.

PHYSICAL PARAMETERS OF NEONATAL CRANIAL ULTRASOUND

The potential for ultrasound to interact with biological tissue has been well established (Shankar and Pagel 2011). The scientific approach to establish safety assurance in diagnostic ultrasound was to consider the possible physical mechanisms that are responsible for biological effects during exposure to ultrasound. The two primary mechanisms involved in ultrasound bio-effects are thermal and mechanical effects. There has been substantial

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research into the thermal and mechanical effects of diagnostic ultrasound on soft tissue and bone, resulting in the implementation of the output display standards (ODS) and the ALARA (as low as reasonably achievable) principle (Barnett et al. 2000).

The ODS assumes linear propagation of ultrasound within a uniform, modestly attenuating tissue and displays thermal (TI) and mechanical indices in real time during the examination (Shankar and Pagel 2011). The TI is the ratio of the current acoustic power output from the transducer to the power required to cause a 1°C increase in temperature (Shankar and Pagel 2011). Three different thermal indices are used to estimate the temperature increase during a scan. These are the TIS, which is applied to exposure of soft tissues; the TIB, which assumes that a layer of bone is present in the focal region; and the TIC, which applies to the presence of bone (cranium) less than 1 cm from the skin (Shankar and Pagel 2011).

The mechanical index describes the relative potential for bio-effects associated with non-thermal mechanisms including cavitation (bubble formation in the presence of gas) (Shankar and Pagel 2011). Clinical safety statements have been published by governing bodies, expressing awareness that pulsed Doppler operates at maximum power outputs and has the greatest potential for biological effects (Barnett et al. 2000). These relate to the routine use of ultrasound for obstetric applications. There are no specific references to neonatal cranial applications. In particular, of the six major bodies that govern the safe practice of diagnostic ultrasound, only the British Medical Ultrasound Society (BMUS) has provided concise and detailed guidelines for the use of ultrasound during neonatal cranial examinations *via* the fontanelle. The BMUS safety guidelines incorporate a limit for the TI in conjunction with the duration of the neonatal cranial scan. They suggest that scanning time be restricted for any TI value >0.7. For a TI of 2.3, it is recommended that the duration of such scans be restricted to 4 min, and scanning of neonatal brain is not recommended for a TI > 3 (BMUS 2010). Although some fundamental research has evaluated the extent of ultrasound-induced heating in the brain, there are no reviews of the literature.

The absorption of energy in tissue leads primarily to a rise in temperature. The temperature elevation produced by ultrasound depends on the spatial peak temporal average intensity (I_{SPTA}), the ultrasound frequency, the dwell time along the beam axis, the width of the beam, the tissue properties, the individual patient and other minor factors.

The interface within the brain itself may be comparable to a fat–muscle or soft tissue–water interface. At these interfaces, only about 1% of the transmitted sound is reflected back to the transducer as echoes (Aldrich 2007). The remaining 99% of the sound energy is attenuated through scattering and absorption (Aldrich 2007). In practice, scattering is thought to contribute a negligible amount of attenuation. Attenuation of the ultrasound beam in soft tissue is a result primarily of the absorption of the acoustic wave motion by tissue, converting its energy to heat (Aldrich 2007; Rumack et al. 1998). The degree of absorption of the acoustic wave in neural tissue will depend on the frequency of the ultrasound beam (Lieu 2010). If the scan frequency is doubled, the attenuation is also doubled. Neonatal cranial scans are typically carried out using high frequencies within the range 10–15 MHz compared with the 2.5–5 MHz that is used for obstetric/fetal applications.

In a recent survey exploring worst-case power outputs as measured in water, the mean I_{SPTA} for B-mode imaging was 341 mW/cm^2, whereas it was 860 mW/cm^2 for pulsed Doppler and 466 mW/cm^2 for color flow Doppler (Martin 2010). Under the assumption of average acoustic and thermal parameters for soft tissue, the temperature rises calculated are given in Table 1 (Ter Haar 2011). In general, these temperature elevations are considered to be over-estimates, as they do not account for thermal conduction, the cooling effects of blood flow or movement of the transducer. However, in narrow, focused beams, such as those used in cranial examinations, the cooling effects of vascular perfusion are negligible (Duggan et al. 2000). Additionally, scanned beams heat less than stationary beams because of the reduced I_{SPTA} (Ter Haar 2011). The temperature rise during many diagnostic ultrasound examinations, such as obstetric/fetal applications, is limited by the use of scanned beams, with any point in tissue being interrogated for only a very short time. However, during neonatal cranial scans, the transducer remains stationary over the fontanelle with very minimal movement. Therefore, significantly elevated temperatures, such as those in Table 1, are more likely during cranial scans, based on the combination of a high frequency, stationary transducer and narrow beam, covering a very small surface area.

Table 1. Estimates of rates of temperature rise in soft tissue for different imaging modes*

Imaging mode	Mean I_{SPTA} (mW/cm^2)	Rate of temperature rise	
		°C/s	°C/min
B-Mode	341	0.048	2.88
Pulsed Doppler	861	0.123	7.38
Color Doppler	466	0.066	3.96

* These assume average acoustic and thermal parameters for soft tissue of absorption coefficient, $\mu_a = 0.06$ neper/cm (0.5 dB/cm) at 1 MHz, and heat capacity, $C = 4.18$ J/g/°C.

Reprinted, with permission, from Ter Haar (2011).

When bone is in the path of an ultrasound beam, the potential for tissue heating is far greater. This is due to the high ultrasonic absorption by bone, which is at least 30 times greater than that for soft tissue (Barnett et al. 1994; Shankar and Pagel 2011). For example, Duck (1990) gives an average absorption coefficient value of 0.6 dB/cm/MHz for brain and of approximately 20 dB/cm/MHz for skull bone. Therefore, the rate of heat deposition at the bone surface is much greater. This is an important consideration when scanning the newborn brain as the transducer is placed directly on the fontanelle between the skull bones. Tissues at most risk from ultrasound exposures are therefore bone, especially developing bone, and any soft tissues immediately adjacent to it, which may experience significant heating through absorption as well as thermal conduction.

The aim of this article was to review the evidence on fetal and neonatal brain tissue heating after exposure to ultrasound.

REVIEW

An electronic search of published papers, abstracts and review articles using Embase, Medline/PubMed, Web of Science and Google Scholar was conducted. All references concerning ultrasound-induced heating of animal and human tissue were identified using a combined text word and MESH heading search strategy with the key words *fetal brain*, and *neonatal brain* and *ultrasound-induced heating*. Publications in languages other than English were excluded. The search yielded 41 related references. All cited studies were based on animal models and all had been approval by a local institutional care and animal use committee and by a local institutional review board.

Over the last 40 y, since the first application of neonatal cranial ultrasound (Canpolat and Yurttutan 2011; Pape et al. 1979; Steggerda et al. 2009), only six studies have published findings on ultrasound-induced heating of fetal skull and brain (Bosward et al. 1993; Carstensen et al. 1990; Duggan et al. 2000; Horder et al. 1998b; Taylor et al. 1998; Vella et al. 2003). However, none of the studies published are reflective of modern ultrasound scan practice.

ANIMAL MODEL VERSUS HUMAN INFANT

Three of the studies (Bosward et al. 1993; Carstensen et al. 1990; Horder et al. 1998b) modeled heating of the fetal brain *in utero* using non-diagnostic ultrasound equipment, but with acoustic output parameters reflective of those used in clinical practice (Table 2). Two of these studies used guinea pigs of 60–66 d gestation (0.9 gestation) (Bosward et al. 1993), and the other study used mature mice (6–7 mo) in a water bath to model fetal exposures (Carstensen et al. 1990). The remaining two studies used neonatal pig (aged 1–7 d) as a model (Duggan et al. 2000; Taylor et al. 1998). Mature mice and guinea pigs (60–66 d gestation) have a cranial bone (parietal bone) thickness of approximately of 0.5 mm, which represents the mean human cranial bone thickness early in the third trimester (Horder et al. 1998a). Term human infants have a skull thickness of 1–2 mm (Li et al. 2015). Hence the bone thickness in all three animal models may be applicable to human newborns with respect to skull thickness, depending on the degree of prematurity. However, all models lack a fontanelle, which reduces the usefulness of these models for human comparisons.

INTRACRANIAL TEMPERATURE ELEVATIONS

The three *in utero* studies discussed above reported mean ultrasound-induced temperature increases of 4.3°C (Horder et al. 1998b), 5.2°C (Bosward et al. 1993) and 5.6°C (Carstensen et al. 1990) at the inner table of the skull, at the level of the bone–brain interface. Approximately 90% of the maximum temperature increase in these studies occurred between 30 and 60 s, reaching equilibrium within 120 s and plateauing thereafter. The temperature rise reported in all three studies is above the threshold recommended by the World Federation for Ultrasound in Medicine and Biology. According to the recommended standards, a diagnostic exposure that elevates fetal *in situ* temperature above 41°C (4°C above

Table 2. Exposure parameters for different studies

Reference	Frequency (MHz)	Power (mW)	I_{SPTA} (W/cm^2)	Beamwidth (cm)	Duration (s)	Mean ΔT (°C)	Animal model	Skull thickness (mm)
Bosward et al. 1993	3.2	260	2.9	0.27	120	5.2	Guinea pig fetus (60 dg)	0.3
	3.2	1120	2.5	0.72	120	15.9		
Carstensen et al. 1990	3.6	1000	1.5	0.28	90	5.6	Mice (6 mo)	0.5
Horder et al. (1998b)	3.5	240	2.8	0.26	120	4.9	Guinea pig fetus (66 dg)	0.4
Duggan et al. 2000	3.5	600	3.6	0.3	90	2.4	Neonatal pig (1–5 d)	0.8
Taylor et al. 1998	7.0	324	0.5	Unknown	300	0.3	Neonatal pig (7 d)	0.8

normal temperature) for ≥5 min should be considered potentially hazardous (Barnett 2001). The objective of the first study by Horder et al. (1998b) was primarily to test the effectiveness of the TI as an estimate of intracranial ultrasound-induced temperature increase. The actual temperature increase in that study was significantly higher (1.3 times) than the value estimated by the TIB (which was 3.3) (Horder et al. 1998b), challenging the estimates produced by the TI as a reliable indicator of intracranial thermal change. The authors suggest that operators should use the TI display with care and err on the side of caution when scanning the neonatal brain. It is interesting to note that after only 120 s of exposure to pulsed ultrasound, the upper value for the TIB calculated in the study by Horder et al. considerably exceeded not only the BMUS TIB limit, but also the recommendations for scan duration. Given that temperature increases plateau after about 60–120 s, we must assume that the elevated temperature reported in those studies is maintained throughout the duration of the scan, further contributing to potential bio-effects.

EFFECT OF BONE THICKNESS ON TEMPERATURE ELEVATION

The two other animal studies used the neonatal pig (age 1–7 d) as a model (Duggan et al. 2000; Taylor et al. 1998). In one of these studies, the reported maximum temperature increase was 2.4°C after 90 s of exposure (Duggan et al. 2000), and the second study reported no statistically significant temperature elevation from cranial ultrasound after a 5-min exposure (Taylor et al. 1998). Although the latter study was conducted under otherwise clinically relevant conditions, both reported a lower temperature change relative to the other studies discussed. This disparity may result from the thickness of the skull of the neonatal pig. Given the high absorption coefficient of bone, the approximate 0.3-mm difference in bone thickness between the neonatal pig model and the guinea pig and mature mouse is sufficient to significantly attenuate the sound beam and alter the resultant temperature rise at the bone–brain interface.

The effect of bone thickness on heating was precisely illustrated by Bosward et al. (1993). In equivalent volumes of brain tissue, they reported a threefold increase in soft tissue heating of the central brain when the brain was insonated after removal from craniums of 60-d-gestation guinea pig fetuses (Bosward et al. 1993). When trying to extrapolate these findings to the extremely preterm human infants (<28 wk gestation), we could assume that the high-intensity ultrasound beam directly enters the soft tissues of the brain *via* the wide fontanelle, potentially resulting in temperature increases similar to those reported in brains of guinea pig fetuses without a cranium. In infants born closer to term, the skull thickness approximates 1–2 mm (Li et al. 2015). This increase in bone thickness may further contribute to the resultant temperature deposition in brain tissue. Other parameters, such as the size of the fontanelle and distance between the skull plates, may also affect the degree of heating in exposed tissue. The literature in this field is limited, and more research is needed to evaluate the effects of clinically relevant ultrasound-induced heating on brain tissue according to skull thickness and fontanelle size.

EFFECT OF TRANSDUCER POSITION ON TEMPERATURE ELEVATION

In four of the five animal studies (Bosward et al. 1993; Carstensen et al. 1990; Duggan et al. 2000; Horder et al. 1998b), the transducer was positioned at a minimum distance of 60 mm from the skull, separated in a water bath to simulate an *in utero* setting. This is an important consideration in a clinical setting because neonatal cranial scans are performed with the transducer in direct contact with the scalp, where the sound beam directly intercepts the dura. There are no tissues to attenuate the sound energy, and therefore, the potential for soft tissue heating is far greater. Given the significant temperature rise (4.9–5.6°C) in three reported studies, it is likely that during a clinical neonatal trans-fontanellar ultrasound, the resulting temperature elevations could further exceed the safety recommendations. The contributing factors in human neonatal cranial scans include the absence of bone in the beam path; the use of a narrow ultrasound beamwidth, where the cooling effects of vascular perfusion are negligible (Duggan et al. 2000; Økland et al. 2011; Vella et al. 2003); and the stationary position of the transducer throughout the examination. Because there are no studies reporting on ultrasound-induced temperature elevations in the human neonatal brain, the degree of correlation between the findings of animal studies and the application of these results to the human preterm infant remains to be resolved. Given the difficulty in carrying out such studies in a human cohort, we need to continue to rely on animal and phantom models to obtain more information on brain/scalp temperature changes during a cranial scan.

LABORATORY EQUIPMENT VERSUS DIAGNOSTIC EQUIPMENT

The results of all animal models indicate that there is a temperature rise at the bone–brain interface above the recommended safety threshold (Bosward et al. 1993; Carstensen et al. 1990; Horder et al. 1998b). However, these studies used laboratory function generators to emit ultrasound pulses at pre-determined intensities. All of the studies that reported statistically significant

temperature elevations (Bosward et al. 1993; Carstensen et al. 1990; Duggan et al. 2000; Horder et al. 1998b) applied intensities (I_{STPA}) at least double the value currently permitted by the U.S. Food and Drug Administration (FDA) in diagnostic ultrasound equipment. Diagnostic ultrasound equipment undergoes ongoing technical changes with emerging new imaging modes and pulse sequencing techniques. These are designed to focus the ultrasound beam to a narrow width to improve axial and lateral resolution to improve image quality (Chiao and Hao 2005). This technique focuses and concentrates all the ultrasound energy to a narrow waist (Martin 2010). Given that cranial ultrasound is performed through a small window *via* the fontanelle with very minimal displacement of the transducer, coupled with a highly focused ultrasound beam, the total energy dissipated in a very small interrogated area may potentially result in intensities higher than the global intensity approved by the FDA. Such infringements of the FDA levels have previously been reported (Martin 2010).

Diagnostic ultrasound technology has significantly changed in the last 25 y, associated with a significant increase in acoustic power (Barnett 2000; Chiao and Hao 2005; Henderson et al. 1995; Lieu 2010; Martin 2010). Average intensities for B-mode imaging increased almost 7-fold from 1991 to 1995. This value almost tripled from 1995 to 2010, which represents a 20-fold increase from the originally published measurements in 1991 (Duck and Martin 1991). In this era of technological revolution and the continuous demand for improved image quality and associated increasing acoustic output levels, it is possible that acoustic safety thresholds are overstepped (Martin 2010). Therefore, the findings of these earlier animal studies that used much higher non-diagnostic levels of ultrasound intensity may not be far from the power levels used in current clinical cranial ultrasound scans and should be considered when assessing heating risks in modern clinical practice.

It is well established that the highest exposure intensities during a scan are associated with pulsed Doppler mode. Therefore, safety guidelines to date have placed greater emphasis on limiting pulsed Doppler use in neonatal applications, with minimal consideration for B-mode imaging, which is used throughout most of the scan. However, ultrasound intensity levels (I_{STPA}) for B-mode have increased at a significantly greater rate relative to those of Doppler mode. In 2010, the mean ultrasound intensity values (I_{STPA}) during pulsed Doppler were approximately 2.5 times higher than those during B-mode, whereas between 1991 and 1998 this ratio varied between 9.2 and 81 (Duck and Martin 1991; Martin 2010). In practice, this means that with modern clinical equipment, B-mode alone could potentially be capable of producing high intensities, especially given the stationary transducer (Duck and Martin 1991; Henderson et al. 1995; Martin 2010). A recent study on *ex vivo* lamb brains reported that 5 min of B-mode exposure using clinically relevant equipment resulted in significantly higher temperature increases on the scalp than examinations in Doppler mode ranging between 15 s and 3 min (Schneider and Lombardo 2016). The role of B-mode in potential heating of the scalp has been largely overlooked to date but warrants more evaluation given the current power outputs.

EFFECT OF ULTRASOUND ON BRAIN DEVELOPMENT

Although temperature elevations have clearly been observed during cranial ultrasound, little is known about associated structural and developmental effects, including functional short- and long-term effects on the developing brain. Two studies examined the effect of ultrasound at a cellular level in animal models (Ang et al. 2006; Liebeskind et al. 1982). Liebeskind et al. examined the effects of pulsed ultrasound *in vitro* at a cellular level using cells of peritoneal fluid of male Sprague–Dawley rats and 3 T3 cells. They found that low-level pulsed ultrasound (15 mW/cm^2 for 30 min) using diagnostic equipment induced changes in the cell membrane, cell surface motility and cellular architecture. The study also found persistence of abnormal behavior and motility in cells 10 generations after insonation, suggesting permanent hereditary effects. However, it is not known whether the *in vitro* effects of ultrasound also occur *in vivo*. Investigations of cells *in vitro* in liquid suspension may be analogous to those in the *in vivo* situation, where the cells are carried in suspension in a fluid such as blood or various transudates. However, the situation in human solid neural tissue might not be comparable. Ang et al. (2006) reported on the influence of prenatal exposure to ultrasound on neuronal migration in the fetal mouse brain. The brains, removed 10 d after birth, manifested no difference in brain size or gross cortical architecture, but there was a statistically significant dose-dependent difference in neuronal dispersion in animals that had been exposed to ultrasound for ≥30 min or more *in utero*.

In neonatal and adult mammals, the neurons acquire their appropriate positions according to a precise schedule and along well-defined and restricted pathways (Altman 1969; Doetsch et al. 1997; Hatten and Mason 1990; Kopen et al. 1999; Lois and Alvarez-Buylla 1993). As a result of this precise schedule, the process of neuronal migration is highly sensitive to a variety of biological, physical and chemical insults (Ang et al. 2006; Barnett 2000; Edwards et al. 1995; Lazebnik and Varich 2007; Liebeskind et al. 1982). Acoustic waves can interfere with the rate of neuronal migration and alter the

sequence of arrival, as indicated by Ang et al., who reported that exposure of the cerebral cortex to diagnostic ultrasound for 5 and 15 min did not yield statistically significant results with respect to the proportion of cells that exhibited altered migration. However, exposure for 30 min arrested 9% of neurons along the migratory pathway. The study also observed a cumulative dose–response relationship, indicating that longer ultrasound exposure times were directly associated with an increase in the percentage of abnormal distribution of neuronal cells. Under the experimental conditions, doubling exposure time from 15 to 30 min increased the rate of abnormal cell migration from 5% to 9%. Exposure longer than 60 min increased the abnormal migration to 11%. Although the length of exposure, the static position of the transducer and the continuous insonation of tissue are not reflective of clinical obstetric practice or neonatal cranial scans, the observed changes in neuronal migration occurred at intensity levels of less than one-tenth (1.5 mW/cm^2) of the FDA limit. By today's standards, the output levels used by Ang et al. are extremely low. This study has highlighted that acoustic energy has the capacity to affect normal anatomic and functional outcomes during brain development. We can therefore not be sure that current clinical exposure is without potential harm, particularly in the very premature infant.

It has been suggested that exposed ectopic cells may develop abnormal synaptic connectivity resulting in behavioral deficits, as has been observed in mutant mice with malposition of neurons (Caviness and Rakic 1978). These ectopic cells can also cause abnormal electrical discharge, which has been associated with epilepsy (Rakic 1988). However, the relationship and clinical relevance of such data to humans is at this point unclear. To conduct such studies on the human preterm brain, one must contend with almost insurmountable problems, because of the difficulty of designing and managing a controlled study, and a range of ethical considerations. We speculate that similar effects may occur in the human neonatal brain with prolonged ultrasound exposure, in particular in very premature infants. However, at this stage, we have very little evidence to provide clear guidelines for clinical practice.

In current clinical practice, the duration of a neonatal cranial ultrasound of the human infant is typically $<$15 min. It is also assumed that the output display standard of the TI is sufficient to provide an indication of the risk of thermal changes for operators, despite reports of its poor reliability (Horder et al. 1998b). However, the mathematical models used to determine the TI do not include any factors associated with the time taken to perform the scan. Given that most of the heating occurs within the initial 30–60 s of exposure and then reaches a plateau, scan duration remains an important component in limiting potential bio-effects.

EFFECT OF ULTRASOUND ON COGNITIVE FUNCTION

Although there have not been any reports on gross structural changes associated with exposure to ultrasound, there exists the possibility that more subtle effects—such as cognitive function—occur, which may remain undetected in experimental studies or in clinical practice. Studies using rodents and chicks as a model have addressed these potential associations (Barnett 1986, 2003; Hande and Devi 1993; Hande et al. 1993; Norton et al. 1991; Schneider-Kolsky et al. 2009; Vorhees et al. 1994). Some of the studies reported that levels of hippocampal noradrenaline, dopamine, serotonin and 5-HIAA, which are known regulators of learning and memory function, were significantly reduced on postnatal assessment in the ultrasound-exposed group compared with controls. The results have highlighted that neurotransmitter regulation, which is vital for cognitive function, was altered by ultrasound exposure *in utero*, even though postnatal growth and physiologic responses remained unaffected into adulthood. However, all of the studies used whole-body antenatal exposures *in utero*. A number of the studies also applied unacceptably large-intensity exposures, and were carried out in very early gestation during organogenesis. These conditions are not reflective of clinical practice.

To date, only one study (Schneider-Kolsky et al. 2009) has investigated the effects of focused ultrasound exposure of premature brains on subsequent learning and memory function. In that study, chicks were exposed to varying durations and modes of ultrasound *in ovo* 2 d before hatching. Chicks were thereafter allowed to hatch naturally, and short- and long-term memory function was evaluated 2 d after hatching. The results indicated that diagnostic levels of B-mode exposure for up to 10 min did not impair memory function. However, short- and long-term memory was significantly impaired after 4 min of exposure to pulsed Doppler ultrasound. Furthermore, exposed chicks were unable to be retrained a few days after the memory testing. Again, it is difficult to correlate the present data with human development. The chick is a favorable model for the study of cognitive impairment because they have well-defined memory stages and because the embryos develop in the egg without maternal and placental influences. However, extrapolation from the chick experiment to humans requires further data from exposures at comparable developmental phases in the two species. Further studies are required to investigate the morphologic damage by ultrasound under various scanning conditions using a

combination of B-mode and Doppler to reflect real clinical practice. Nevertheless, the results add to the pool of animal studies, highlight the possibility of adverse effects under prolonged exposure conditions and emphasize the need for practitioners to be aware of dwell times of the ultrasound beam, particularly during pulsed Doppler.

There is a paucity of data and a lack of recent studies concerning ultrasound-induced heating of neural tissue, especially under clinically relevant conditions that emulate the neonatal cranial ultrasound of the human neonate. To provide more robust evidence for heating effects of neonatal ultrasound in humans, animal studies need to consider the appropriate size and gestation of the human neonatal brain, as well as the thickness of the skull of the preterm human infant, the use of clinically relevant acoustic outputs from modern diagnostic equipment and the position of the transducer, which is in direct contact with the dura, to appropriately reflect the scan of the preterm human brain (*i.e.*, Not *in utero*).

CONCLUSIONS

There are methodological limitations in the majority of the animal studies. Nevertheless, the results of the animal models suggest that ultrasound can result in temperature rises at the bone–brain interface above the recommended safety threshold. More research is needed to elucidate the structural and functional effects of clinically relevant ultrasound using modern ultrasound machines.

Safety recommendations are based on the results of outdated studies carried out 25 y ago. These do not account for the effect of technological advances that have seen dramatic increases in the diagnostic power of modern ultrasound equipment. Scan duration remains the most important component in limiting potential bioeffects, but this is not yet recognized by five of the six major professional bodies that govern the safe practice of diagnostic ultrasound. In the meantime, operators are advised to limit the use of Doppler mode, as well as reduce the overall duration of the neonatal cranial scan.

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