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7 Biological Effects in Laboratory Animals

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In animal studies, attention has been given to a number of ultrasonically induced biological effect end-points. One can think of these as being classified into one of two general categories: structural or functional alterations. A change in biological material that is determined through histologic means is considered a morphologic or structural alteration. Most ultrasonically induced structural alterations have been assessed by light microscopy. A biological effect which is assessed by a change in some biochemical level, pH, or activity is considered a functional alteration. In general, relatively high ultrasonic intensity levels are required to produce a structural alteration; at lower levels, where structural alterations are not detectable, functional alterations have been observed.

In the context of a structural or functional alteration, there are various degrees to which the experimental data are conflicting. One category of observations deals mainly with structural alterations (usually termed "lesions" here) of biological tissues produced by quite high levels of ultrasonic energy. Here there is no conflict in terms of whether or not a specific effect occurred but there are conflicting viewpoints in terms of the fundamental mechanism or mechanisms responsible. At lower ultrasonic energy levels, usually within the therapeutic range, there are conflicting viewpoints as to whether, or to what degree, a structural alteration occurred. And, for a third general category, at ultrasonic energy levels lower than the therapeutic range, sometimes at diagnostic levels, there are no indications of structural changes and there are very conflicting data as to whether a functional alteration occurred.

HIGH-INTENSITY ULTRASONIC STUDIES

Some 35 years after the Curies discovered piezoelectricity in 1880,[1] the French scientist Paul Langevin developed the first use of ultrasonic energy wherein underwater acoustic echoes were bounced off of submerged objects.[2,3] During the course of this work, the first reported observation was made that ultrasonic ally weighed. The data showed a statistically significant fetal weight reduction from about 6 to 18 percent, depending upon the exposure conditions. There were seven exposure groups, including a sham. Two hundred and seventy-two litters (2,866 fetuses) were exposed under conditions ranging from 0.5 to 5.5 W/cm^2 for the spatially averaged intensity (I) and from 10 to 300 seconds for the exposure time (t). A dose-effect dependence of exposure condition versus average fetal weight was examined by defining the dose parameter I^2t.[37]

The observation that in utero ultrasonic exposure of mice can cause weight reduction in fetuses compared with the sham has also been confirmed by two other groups using two different strains of mice, namely, LAF$_1$/J mice[38,39] and CFW Swiss-Webster mice.[40] In the earlier study,[38] relatively high-level pulsed ultrasound conditions (center frequency around 1 MHz) were employed, and a significant reduction in fetal weight was reported for SPTA intensities above 50 W/cm^2 [maximum intensity (I_m) of 2,936 W/cm^2] and exposure times of 20 seconds when fetuses were irradiated on the eighth day of gestation. The later report by Fry et al.[39] indicated that the highest exposure conditions [maximum intensity (I_m) of 1,936 W/cm^2, SPTA intensity of 51 W/cm^2, and exposure time of 20 seconds] produced a statistically significant 18.8 percent fetal weight reduction (relative to the sham animals), whereas at a SPTA intensity of 45 W/cm^2 and lower [maximum intensity (I_m) of 1,936 W/cm^2 and exposure time of 20 seconds], there was no change in the fetal weight relative to the sham. Stolzenberg et al.[40] reported on fetal weight reductions ranging up to 25 percent relative to the sham animals when the mice were ultrasonically exposed (CW 2 MHz, spatial-average intensity of 1 W/cm^2, exposure times up to 200 seconds) at gestational ages of 0, 7, or 12 days and examined on the 17th day of gestation.

A preliminary study conducted at even lower ultrasonic levels suggested that the fetal weight reduction may be sustained postweaning.[41] Time-mated CF$_1$ mice were irradiated at the 13th day of gestation with CW (1 MHz) ultrasound and examined at 55 days postconception (approximately 2 weeks postweaning). There were three exposure groups (sham, 0.25 W/cm^2, and 0.80 W/cm^2 spatial-average intensity, each for 120 seconds) and the data yielded statistically significant weight reductions of 8.7 percent and 14.8 percent, respectively, relatively to the sham. However, in a follow-up study, Stratmeyer et al.[42] did not confirm this earlier finding of sustained postweaning weight reduction. Rather, a weight gain compared to the sham was suggested. Fetal weight reduction was not observed in rats exposed on the ninth day of gestation to CW (3.2 MHz) ultrasound, even at exposure conditions that produced some mortality.[43] There were, however, a few pups stunted but this observation was not statistically significant.

For further discussion of fetal studies, see Chapters 8, 9, and 10.

CONCLUDING COMMENTS

An important concern deals with the question of the significance of an ultrasonically induced biological alteration, for example, as viewed with respect to risk. But very little effort has gone into the assessment of specific biological alterations. This is true, in part, because appropriate dose-effect responses of these alterations have not been developed. Rather, a single dosimetric condition has been utilized for the experiment and this has precluded our ability to investigate the role of that particular biological alteration in terms of extrapolating it from the experimental animal to human beings. Herein lies the greatest importance of investigating ultrasonically induced biological alterations in experimental animals. Such biological observations can provide insight into the risk, if any, to which humans might be subjected. Dose-effect observations provide the fundamental basis from which mechanisms of interaction can be determined and thus provide a firmer scientific basis for extrapolation to humans.

In conclusion, based upon experimental animal studies, it appears that the available information suggests that the risk associated with the clinical use of ultrasound is quite low. However, our knowledge regarding ultrasonic bioeffects and biophysical interaction is rather incomplete at this time. Because of this apparent paradox, it is essential for the clinicians to be provided with up-to-date information on potential risks so that they can continue to render an informed benefit-risk judgment. The principal source of such data is from animal experiments. It is difficult to evaluate any of these studies in isolation, especially when there are conflicting observations. And there will be conflicting observations especially as the ultrasonic levels decrease. This is a given and it must be understood.

ACKNOWLEDGMENTS

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