The Need for Sound and Vibration Standards in U.S. Research Animal Rooms

The impact of research animal exposure to noise and vibration in vivariums and animal housing is a frequently overlooked aspect of biomedical research, though there is emerging recognition and study of its significance.

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The research animal housing and life sciences laboratory industry in the U.S. is not currently governed by accepted standards for acoustics or vibration. This is probably one of the last sets of environmental influences (along with CO_2 and ammonia levels) that have yet to be given serious consideration.

We are playing catch-up to our European colleagues in this regard. Among European standards for individually ventilated cage (IVC) racks are those based on accepted IVC testing parameters developed by the Center for Biomedical Research of the Faculty of Medicine (TierschutzInformationsZentrum für die Biomedizinische Forschung der Medizinischen Fakultät: TIZ-BIFO), at Ludwig-Maximilian University of Munich. TIZ-BIFO testing assesses IVC reliability, safety, and quality through tests of ventilation, climatic and acoustic parameters, as well as factory inspections and field acceptance tests of IVC installations.

In the absence of U.S. standards, Allentown and other IVC manufacturers have achieved TIZ-BIFO standards in the interest of helping shape future U.S. acoustical standards and to provide customers with statistical data which proves the efficiency and safety of our acoustical design parameters as it relates to reliable research outcomes and to animal exposure.

Changing Animal Models

Our industry has seen significant change over the last 20 years. The animal models that we use now are very different. Previously, research involved larger animals that don't experience noise stress to the level that rodents do.[48] Until facilities actually began to house large numbers of rodents, and use ultrasonic frequencies and ultrasound-generating equipment in proximity to them, the industry wasn't as concerned about sound, especially in the frequencies outside the range of human hearing. Now, with the changes that have taken place, concern is growing because of the potential negative impact sound and vibration can have on research paradigms.[19, 28, 35, 40, 48, 52]

As a foundation, we define our subject matter as:

Sound – mechanical radiant energy that is transmitted by longitudinal pressure waves in a material medium (such as air) and is the objective cause of hearing.

Noise – any sound that is undesirable or interferes with one's hearing of something.

Vibration – a periodic motion of the particles of an elastic body or medium in alternatively opposite directions from the position of equilibrium when that equilibrium has been disturbed (as when a stretched chord produces musical tones or particles of air transmit sounds to the ear).

From the IVC manufacturer's standpoint, a customer's perception of the loudness or quietness of a cage rack is highly subjective. Hard scientific data is needed about threshold frequencies and sound pressure (decibel) levels stressful to rodents that can be taken into account in the design cycle so that these issues are addressed. Aside from individual manufacturers' internal research, which is largely proprietary in nature, there is very little data involving planned studies over wide frequency ranges and different sound pressures.

Ultrasound: An Emerging Factor

The advent of new technologies compounds the necessity of investigating sound, frequency, and vibration levels. We know of anecdotal evidence where ultrasound from a personnel sensor in a vivarium airlock produced seizures in mice which investigators were taking through the airlock. Or consider the ultrasound generated by the ballast in an overhead fluorescent light fixture. We are aware of facilities where that is enough of a concern that fluorescent ballasts are being placed in a remote location outside the animal housing room. These are good examples of how much more we need to know through planned scientific study.

While the industry is beginning to take these issues seriously, the previous lack of interest may stem from the simple fact people can't hear in the same range that animals do, so sound and noise impacts have tended to be ignored. Compared to the human hearing range of 20Hertz (Hz) to 20,000Hz, a rat's hearing range is 200Hz to 76,000Hz, and a mouse's range is 1,000Hz to

91,000Hz.

Ultrasound is any sound frequency above the human hearing range of 20,000Hz. Any frequency below 20Hz is infrasound. We know much of the equipment and ventilation in a vivarium may create ultrasonics that the animals can hear, and which we can't, but this is an area that requires more measurement and study.[34, 44]

The Negative Effects of Noise

Noise exposure can have various negatives effects including:

- Induce or accelerate hearing loss in mice confounding studies involving learning or hearing acuity.[13, 16, 29, 48]
- Change neuroendocrine and cardiovascular function.[5, 18, 19, 23, 24, 28, 38, 48, 50]
- Disturb sleep-wake cycle.[19, 41, 42, 48]
- Induction of seizures in susceptible strains.[11, 12, 26, 27, 48]
- Changes in reproduction and development.[15, 17, 33, 51, 52]
- Alter immune function.[2, 3, 6, 30, 46, 47, 49]
- Alter the toxicologic properties of certain agents.[19]
- Alter weight gain.[35]
- Induction of an array of behavioral changes.[48]
- Cause physical injury due to startle reactions.[22, 25]

Sudden impact noise which is excessive or loud may have greater negative impact than noise that is constant to which animals may adapt. Examples of sudden impact noise include fire alarms, overhead speakers, loud conversations, and equipment collisions. Sudden impact noise induces the startle response and may negatively impact rodents more than other species in the animal facility.[12]

The idea of playing background music as white noise to mask sudden noises in a facility has been around for some time. There is evidence both in support of and against this practice. Intercom systems playing elevator music unquestionably dampen startle response to sudden noises,[37] but there is also evidence that rodent hearing doesn't fully develop normally when subjected to constant white noise, which would argue against this approach.[48]

The negative impacts of auditory stress are not insignificant. For example, banging of cages in an animal room can cause a 100% to 200% increase in plasma corticosterone in rats, which persists for two to four hours.[4] Exposure of pregnant rats to an 85 decibel (dB) to 90dB fire alarm bell results in alteration of immune function in the offspring.[46]

Additionally, there is a fairly sizable body of publications indicating that certain sounds, certain sound pressures (decibel levels), and certain frequencies can be teratagenic, or cause abnormal development of the fetus, fetal malformation, even fetal death in certain species.[33, 34, 46, 48]

Sources of Noise in the Animal Facility

Noise in an animal facility can come from various sources including:

- Ventilation systems
- Operation of equipment
- Husbandry and cleaning procedures
- Vocalization and activity of animals
- Personnel and equipment movement
- Light fixtures and computer terminals

It appears that most noise in the facility results from two sources — the direct activity of people working in the facility and increased activity of animals (vocalization, cage rattling, or banging) in response to the presence and actions of people. Animal facilities are generally quiet when people are not present.[31]

Noise levels in rodent rooms at facilities we have worked in, equipped with individually ventilated cages with rack mounted supply blowers and rack exhaust connected to house exhaust, were typically 50dB to 55dB with the work station off and 70dB to 75dB with the work station on. By way of comparison, conversational speech registers at about 60dB. There was very little difference between micro-environmental and macro-environmental noise levels.

Perkins and Lipman investigated three commercially available IVC systems. All three were shown to produce macro- and micro-environmental noise significantly greater than room background noise. Macro-environmental noise ranged between 74dB and 80dB, while micro-environmental noise ranged between 79dB and 89dB.[39]

Perkins and Lipman did not measure ultrasonic frequencies in their studies but Clough, *et al.* reported no detection of ultrasonic frequencies produced by a ventilated caging system.

Infrasound Sources and Animal Effects

Infrasound is sound of a frequency range below the level of normal human hearing (i.e. less than 20Hz). Infrasound has a relatively long wavelength with a low material absorption rate and thus has the ability to travel vast distances.

Infrasound has an intrinsically mysterious effect as it usually is felt but not heard.

Sources of infrasound include:

- Ventilating systems

• Electric generating plants
• Compressors
• Cooling towers
• Overhead motorway bridges

A study involving the exposure of rats to infrasound for three hours per day for five to forty days resulted in the development of irreversible alterations in the liver characterized by ischemic areas with morphologic and histochemical changes in hepatocytes.[36]

Exposure of guinea pigs and rats to infrasound resulted in changes in the myocardium including spasms of the main coronary vessels which led to the development of ischemia resulting in destruction of myocardiocytes.[1]

The Impact of Vibration

Vibration and sound are very similar; they're just at different frequencies. In fact, sound is vibration. There is a large volume of data that documents the harmful effect on humans due to occupational exposure to excessive vibration.[7-10, 14, 20, 21, 45] This data may be applicable to animals, as far as the threshold of perception, i.e., at what specific level vibration becomes unpleasant and at what level it becomes intolerable. Again, research is needed, for we know even less about vibration than we do about sound.

There are certain vibration sources that cannot be controlled. Any building that is near a subway experiences vibration when the trains pass through. We know that when there is major construction adjacent to or near rodent housing facilities, we commonly see a reduction in breeding efficiency and an increase in behavioral aberrations in the animals, which is probably induced by the stress of vibration from the construction.

The fixed mechanical equipment in most large research buildings is also a source of vibration which, in a vivarium facility, the building's design should attempt to isolate.

Aside from external factors, vibration sources in the animal holding room are primarily equipment-based — racks, blowers, and work stations. In the absence of U.S. standards for vibration in animal rooms, there are opportunities for equipment manufacturers to participate in third-party testing to generate data which proves that their equipment is not a significant source of vibration. Allentown and other IVC manufacturers have conducted such third-party verification and the results may be useful in establishing eventual vibration standards.

Among the anecdotal impacts of vibration are a reduction of breeding efficiency in rodent breeding colonies, reductions in food intake and weight gain, and behavioral modifications.

A controlled study in which mice were subjected to a simulated severe earthquake and five aftershocks found a very significant increase in the rates of cleft palate and fetal resorption.[32]

In another controlled experiment, rats subjected to vibration amplitudes of 4.6cm at 283 cycles/minute for 15 to 30 minutes a day for 21 days, exhibited severe effects on body weight, food consumption, leukocyte counts, and organ weights.[43]

A Holistic Approach

It becomes evident that minimizing the impact of sound and vibration requires thinking beyond the IVC to everything that is in the macro-environment of the animal room or that integrates with the room. Every piece of equipment in the room or external system that touches the room should be reviewed.

There has been increased focus recently on greater equipment integration into the building such as taking the supply and exhaust blowers off the cage racks and putting them in the interstitial space above the ceiling to get noise, vibration, and heat load associated with the blowers out of the room.

Installing noise-abating facility air ducts instead of standard hard ducts can provide additional and significant noise mitigation. We have experience with a facility which reduced the sound level in the rodent room from 70dB to 50dB simply by replacing hard ducts with noise-abating ductwork.

There is other equipment that contributes to the noise level of the room. Laminar flow work stations, when operating, can add about 20dB to the noise level of the room with the ventilated racks typically running at 55dB to 60dB. Moving the work stations to another room is a potential solution.

Room construction materials shouldn't be overlooked. Surfaces such as walls, ceilings, and floors are typically smooth, impermeable, sanitizable materials which are sound-reflective, not sound-absorbent. Personnel may take computers into the room. Computer monitors have a high ultrasonic emission.

To create a truly quiet environment for animals, we must look beyond the rack to the construction of the room, other equipment in the room, and any penetration into the room such as HVAC or monitoring equipment. Nothing can be ignored.

Towards Sound and Vibration Standards

Clearly, there is a real need for scientifically-based sound and vibration standards, though the research community is probably in a position to set standards to some degree just based on empirical knowledge. We are aware of architects involved in the design and construction of vivariums who recommend 45dB as a standard for the rack and the room. In the absence of sufficient hard data, 50dB seems an appropriate level given the empirical evidence.

The industry needs to study rodents by generating noise at different frequencies and sound pressures across those frequency ranges to look for stress response in the animals through behavioral-based observation or by actually measuring levels of stress hormones, or simple central nervous system activity in sound-responsive brain areas. This is a fairly complicated issue because rodents hear across a broad sound frequency range. Yet, if studies could determine the specific sound and vibration level limits, standards could be established that would be valuable to the industry at large.

One of the challenges in investigating these issues is that it is very difficult to get funding to generate this kind of data. Manufacturer testing therefore is truly important. Additionally, the AALAS Foundation may be a source of funding available for these types of studies which, again, benefit the entire industry.

On another front, the Institute of Noise Control Engineering, an international professional organization whose members have expertise in engineering solutions to environmental noise problems impacting human hearing levels, has expressed interest in working with experts in the biomedical community to develop standards for laboratory animals. The INCE has worked in the past with organizations and experts in other industries on noise issues to set and publish standards.

Were we able to develop standards through such a partnership, the marketplace would drive acceptance of them as a marketing and funding differentiator for manufacturers and facilities who could prove performance and research advantages within the specified parameters.

While a part of normal daily life, noise and vibration may significantly impact biomedical research and steps should be taken to minimize their effects. Given a clear need and clear benefits, developing sound and vibration standards for animal rooms would advance the quality of the equipment and ensure the quality of research in the biomedical research industry.

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