In Utero Low Frequency Noise Exposure in Rats - II

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Introduction This report focuses on occupational exposure to low frequency noise (LFN) (≤500 Hz, including infrasound). Long-term (years) LFN exposure can lead to the development of vibroacoustic disease [1]. Pregnant women often carry their pregnancies to term while working in LFN environments. The general population can also be exposed to LFN in a variety of common, everyday activities. The purpose of this study is to describe the effects of LFN on the respiratory epithelia of rats exposed to LFN in utero and subsequently exposed to additional LFN.

Methods Animals. Ten Wistar rats were gestated and born while exposed to LFN on an occupationally-simulated schedule: 8 hrs/day, 5 days/week, weekends in silence. Control rats were gestated in equal living conditions, but in continuous silence. All animals were fed standard rat food, had unrestrained access to water, and were treated in accordance with applicable legislation (86/609/CE). After birth, animals were exposed to an additional 235, 2213, 4399 and 5304 hours of LFN. Noise Exposure. The overall linear and A-weighted noise and spectral analysis collected inside the rat chamber using a digital real time analyzer (B&K 2144). An analog noise generator produced an amplified and frequency filtered acoustic signal so that acoustic energy was highly concentrated in the lower frequency bands, 50 Hz to 500 Hz, exceeding 90dB LIN. The overall linear sound pressure level was above 109dB LIN, and the A-weighted level was around 98dBA. Microscopy. After birth, animals were exposed to additional LFN in varying amounts, then removed from the LFN environment, and sacrificed by a lethal intravenous injection of sodium-pentobarbital. The trachea was divided in two, along the saggital line. Specimens prepared for scanning electron microscopy (SEM) (JEOL JSM-35C) were dehydrated, critical point-dried, coated with gold-palladium and examined with the electron microscope at an accelerating voltage of 15 kV.

Fig. 1. SEM of rat gestated in LFN + 235 hrs of LFN. Rosetta-structure centered on a BC (arrow) is readily identifiable. BC microvilli are clusted together and surrounding cilia are shaggy.

Fig. 2. SEM of rat gestated in LFN + 235 hrs of LFN. Depleted ciliary fields and a sheared cilia (small arrow). A BC (large arrow) with abnormal microvilli surrounded by SCs. A sunken, or necrotic, BC (arrowhead).
**Results** Ciliary populations were visibly shaggy, and sheared cilia were frequently observed. SC microvilli were shorter than in controls, and appeared stunted. BC microvilli grouped together, forming regular clusters of microvilli, and losing the uniform distribution seen in controls. With increasing exposure time, BC microvilli fused together. After 4000 hours of LFN exposure, rosetta-structures became difficult to identify.

![Fig. 2. SEM of rat gestated in LFN + 2213 hrs of LFN. Ciliary fields are depleted. BCs (arrows) are surrounded by rings of SCs. Intercellular junctions are thick and prominent.](image)

![Fig. 4. SEM of rat gestated in LFN + 5304 hrs of LFN. Sunken BC (arrow). Some individual microvilli are seen around the edges. Isolated strands of cilia are visible.](image)

**Discussion** The reduction of ciliary populations is consistent with what was previously observed in LFN-exposed rats not gestated and born in LFN [2]. Aspects of cellular de-differentiation were also seen in other LFN-exposed rat studies [2]. The genotoxic effect of LFN, as measured by the frequency of sister chromatid exchanges, has been previously demonstrated in both human [3] and animal [4] models. Thus, metaplastic cellular organizations are not entirely surprising. Moreover, in vibroacoustic disease patients who suffered respiratory tract tumors, all were squamous cell carcinomas [5]. In Wistar rats, the consequences of in utero LFN exposure are real, detectable, and severe. The implications for human fetus’ of LFN-exposed pregnant females are unknown, but results herein vigorously suggest that they should not continue to be ignored.

**Keywords:** vibroacoustic disease, cilia, brush cells, microvilli, noise exposure, occupational

**References**


