Cells and Sound

by Maria Anna Pabst PhD

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Based on the studies of cell biologist Maria Anna Pabst PhD, hypotheses can be formulated of 'vitalising "effect of sound massage - at the cellular level.

Through the Peter Hess-Sound Massage many people have already experienced the beneficial effects of sound on the human body. This raises the question of whether the entire person is necessary in order to benefit from the sounds of a sound treatment that is whether the human psyche is the key factor for the effectiveness of sound massage, or if the sounds already works at the cellular level . With this question in mind, we have examined the extent of the effect the sound of a singing bowl has on cells in a cell culture.
The human endothelial cells were used for our sound experiments. Endothelial cells line the inside of the blood and lymph vessels with a layer of flat cells, i.e. they directly border on the current flowing in the blood vessels or lymph. Those cells in the vessels can certainly adapt to changing mechanical or physiological conditions. Lipton (2006) describes that in his research, endothelial cells in a cell culture, just "watch" their environment and change their behavior according to the information available to them. It has been found that these sensible cells move in towards nutrients and withdraw from toxins. An "intelligent" performance of single cells.
In our experiments endothelial cells were isolated from blood vessels (arteries) of human placenta, placed in culture dishes with a special low-nutrient medium and cultured. Five experiments were carried out with multiple measurements. The cells were each sonicated with a Peter Hess ® Sound Therapy bowl, for one hour on three consecutive days. Type heart bowl (Fig. 1).

Figure 1: Sound of endothelial cells in a cell culture vessel.

This cell culture dishes were covered with a few layers of cellulose and the singing bowl positioned on top. The singing bowl was touched with a Felt Mallets every ten seconds, alternately three times on the left side and three on the right side.
The heart singing bowl used had a diameter of 23 cm. Frequency measurements of the singing bowl and the cell culture dish below were performed with a laser vibrometer and showed a wide range of frequencies that overlap in part and interfere with each other. The data were represented up to 10 kHz.
The main bands of the sound bowl were 455-3472 Hz (Fig. 2a) and in the cell culture dish 442-3421 Hz (Fig. 2b). Beats could be measured only for the sound bowl, but not in the cell culture dish. These sounds come through similar frequencies, both periodically strengthen and extinguish. Beats in the sound bowl led to a periodic volume modulation with a frequency of 5.8 Hz
As a control, in addition to the sonicated cells, cell culture dishes supplied with the same number of endothelial cells from the same isolation, were placed in an adjacent room, and a heart singing bowl placed in the same manner.
The cell culture dishes were allowed to stand for one hour in the same room temperature conditions, without resounding the singing bowl.
One day after the last treatment, the sonicated cells and the cells of the control

were microscopically examined with a phase contrast microscope.
Here, no morphological differences between the cells treated with the singing bowl and the control group were found. A portion of the cells were prepared for electron microscopy to investigate any ultrastructural changes in the cells.

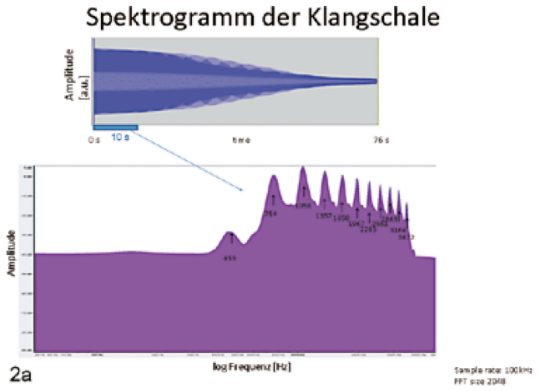

Abb. 2a: Frequenzmessung der für die Beschallung verwendeten Herzklangschale.

Figure 2a: Frequency measurement of the heart singing bowl used for sonication.

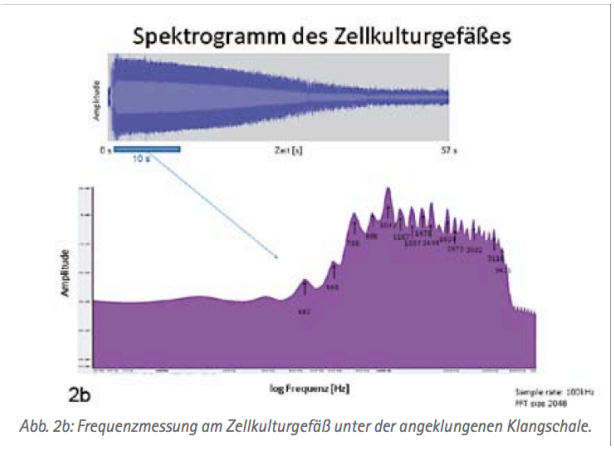

Abb. 2b: Frequenzmessung am Zellkulturgefäß unter der angeklungenen Klangschale.

Figure 2b: Frequency measurement on the cell culture dish under the sonication with singing bowl.

For scanning electron microscopy (SEM observation of surfaces), the endothelial cells were given the opportunity to grow up in the culture dishes on small glass plates. For transmission electron microscopy (TEM, irradiation thin layers) plastic sheets were introduced for growing the cells in the culture dishes. On one hand, it was interesting to see the surface structures of cells in the SEM, on the other hand, it was also important to look with the TEM inside the cells. For this purpose, the cells were embedded in resin and we made about 60 nm thick sections, which allow an assessment of the various cell organelles, which were small organs with different functions in the cells. Endothelial cells form, as already mentioned above, lining the interior of the vessels in the body through a layer of cells. When they are isolated, they attempt to form again in the culture a uniform and closed layer by cell divisions and growth.
During their growth in cell culture, these cells first form out extensions, through which they establish contacts with neighboring cells, to finally respond with further growth with these tight connections. No differences between sonicated cells and control cells were observed in these extensions (Fig. 3a and b). The fine projections on the surface of the cells through the medium (microvilli) showed no morphological differences between the two cell groups. In thin section TEM no differences in the morphology of cell organelles were also detected (Fig. 4a and b).
Nuclei (main carrier of genetic information), rough endoplasmic reticulum (places of protein synthesis), Golgi apparatus (place of processing of proteins and formation of secretion vesicles) and lysosomes (the digestive organelles used for degradation of received or in the cell itself no longer used materials) showed their usual structures. Both the sonicated cells as well as in controls intact cell organelles and relatively common intracellular degradation processes were seen.
In addition, the cells were examined using a Casy cell counter. This is a device, where cells are passed through a fine capillary (glass tube) and individually counted. In addition, the resistance that comes into existence from the charge of the cell membrane can be measured. The charge of the cell membrane provides information on the state of vitality, that is about the "health" of the cells. With the Casy cell counter the number of cells, the number of living cells and the amount of

cellular debris was determined. Also in the nutrient medium, the enzyme LDH (lactate dehydrogenase) was measured, which provides information about the amount of dead cells. This enzyme is usually only available inside cells. When cells perish, LDH is released from them into the surrounding medium, which can then be measured.

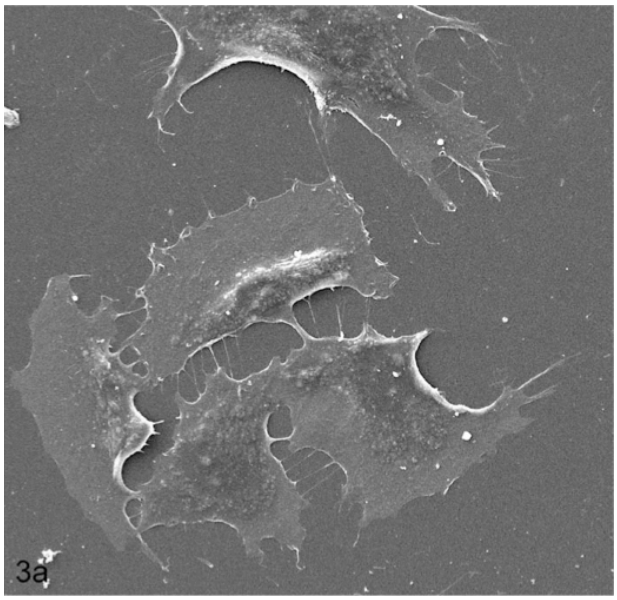

Figure 3: SEM image of sonicated endothelial cells. Original magnification x 750.

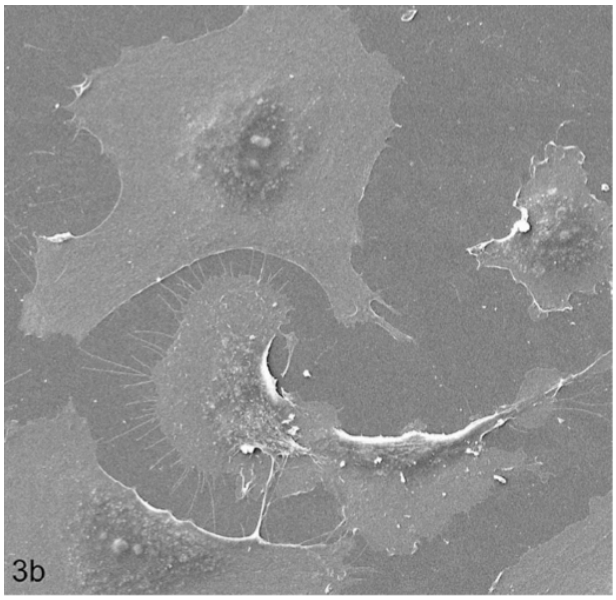

Figure 3b: SEM image of non-sonicated endothelial cells (control). Original magnification x 750.

Although no morphological differences between sonicated cells and controls were found, we found clear differences between sonicated cells and the controls with the Casy Cell Counter. It showed that the total cell number after sonication and the number of living (viable) cells compared with the controls (the latter is set to 1) are significantly higher ($p = 0.026$ and $p = 0.017$) and the amount of cell debris after sonication approximately equal as in the control. The LDH concentration is slightly, but not significantly reduced to the sonication(Fig. 5). Overall, the leaves to the statement that the cells divide more after sonication and the death rate is slightly reduced.

What's going on in the culture medium or in the cells when sound waves interact with them? The culture medium is an aqueous phase and the cells are to a considerable degree water. Water can, when you put it to vibrate at different frequencies, form different sound figures and patterns (Lauterwasser 2002, 2005), and one can create different sound patterns of the water after playing CD's recorded with different music.

Lauterwassser describes that the vibration applied to the water creates a rhythmic motion. This could have an effect on the endothelial cells, when they are brought into vibration by sonication.

In addition to the water element the cell membranes may also play a role in the vibration exposure. On the surface membrane and in membranes in the interior of cells there are variety of proteins are located (protein molecules) with various functions. For example, receptor proteins act as sensory organs (such as eyes, ears, taste organs). Lipton (2006) suggests that they function as molecular nano-antennas that are targeted to specific environmental signals. Those receptors are formed for each environmental signal that they can read. Some receptors respond to physical signals, such as different molecules which are histamine, estrogen and insulin.

According to Lipton, the antennas of the receptors can also receive vibrational energy fields such as light, sound and radio waves. In this case, the charge of the

modified protein and the receptor changes its shape (Tsong 1989).

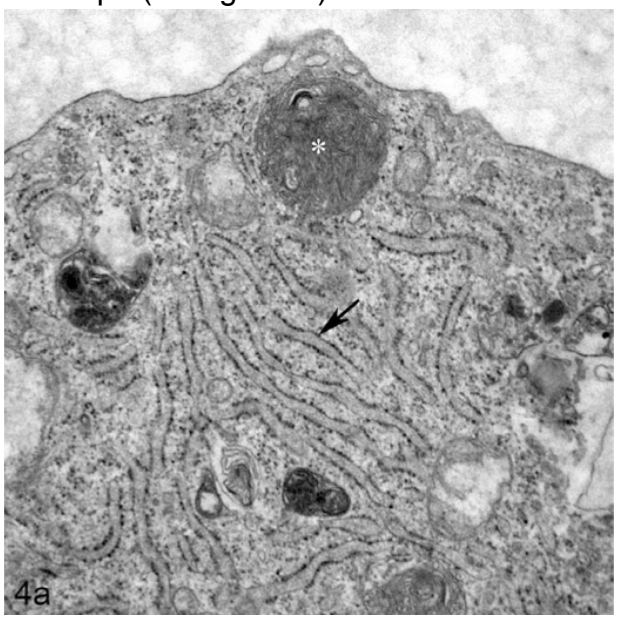

Figure 4: TEM image of a sonicated endothelial cell (cell extract). Rough endoplasmic reticulum (arrow), with lysosome degradation products (asterisk). Original magnification 12,000 x.

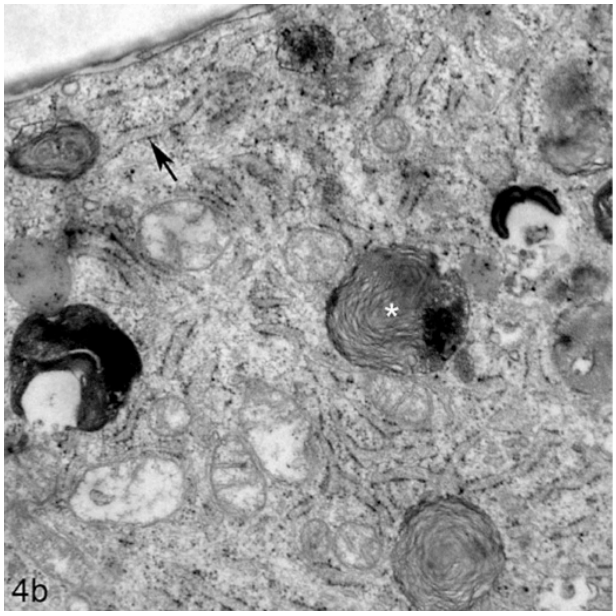

Figure 4b: TEM image of a non-sonicated endothelial cell (cell extract). Rough endoplasmic reticulum (arrow), with lysosome degradation products (asterisk). Original magnification 12,000 x.

Some cells have even specialized in the perception of vibrations. So for example auditory cells have specialized in the perception of mechanical stimuli (sound waves), photoreceptor cells in the perception of electromagnetic waves (light). Also in these cells, the cell membrane plays an important role in the perception of these signals. Thus, receptor molecules allow perception of the environmental signals but also the cell must be able to respond to these signals. , Other proteins are required again, for the setting in motion a reaction mechanism for the environment signals that are processed inside the cell and translated in cell behavior. Why should the endothelial cells that are exposed to the organism at different blood flow currents, not be able to perceive and respond to vibrations?
The singing bowl vibrations transmitted through the cell culture medium not only reach the surface of the cell membrane but are also transmitted to the membranes present in the cell interior so that the entire "organ system" of the cell is in oscillation. The vibrations of the singing bowls not only have an impact on the liquid in the cell area but probably on the membrane system of the cell with its diverse functions. Possibly an environmental signals for the cells also have an impact on processes in the cell nucleus, eg on cell division of "normal" cells, as you can tell from the results of sound experiments.

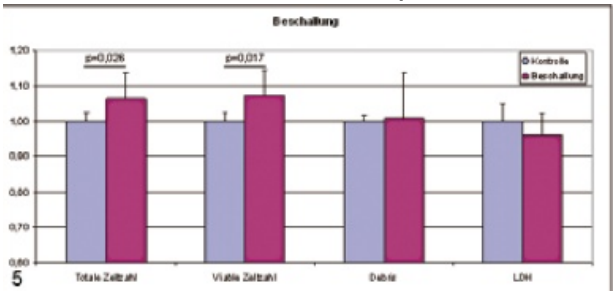

Figure 5: Graphical representation of different measured values of sonicated cells compared to non-sonicated cells (control).

Furthermore, it is conceivable that even natural oscillations of different molecules in the cells affected by the sounds of the bowls, and thus influence is exerted on the cell function.
According to the results described above, the sound vibrations of the bowl appear to

have an activating effect at least to the cell division of the endothelial cells in culture.

The above cell culture experiments were performed at the Institute of Cell Biology, Histology and Embryology, Medical University of Graz in collaboration with Prof. Berthold Huppertz PhD, Prof. Ingrid Lang-Olip PhD, Elisabeth Bock, Angela Schweizer- Trummer MSc and Julia Konig MSc, the frequency measurements at the Institute of Zoology, Karl-Franzens-University of Graz Univ. Prof. Heiner Romans PhD and Manfred Hartbauer PhD.

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Maria Anna Pabst PhD is a professor of Cell Biology, Histology and Embryology at the Medical University of Graz. She is also a meditation leader, has completed training in self-healing methods, and is trained in the Peter Hess-Sound Massage. She works with Katathym imaginative methods (internal pictures), is a Reiki Master and is interested in medicinal plants and natural cosmetics.

Translation : Alex Andrzejewski