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*Original Contribution*

# APPLICATION OF FOCUSED ULTRASOUND FOR THE STIMULATION OF NEURAL STRUCTURES

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**Abstract—The feasibility of the use of focused ultrasound for stimulation of the superficial and deep-seated receptor structures of humans and animals are presented in this review article. Applications of this method in physiology, for research into somatosensory and hearing perception, and also in clinical medicine for the diagnosis of neurological, dermatological and hearing disorders involving changes in perception of sensations different from normal, are discussed. It is proposed that the main effective factor of focused ultrasound as a stimulus of neural structures is a mechanical one. Such a mechanical effect could produce a change in membrane potential resulting in the stimulation of neural structures, which is related to the origin of tactile, thermal and hearing sensations. The direct action of sign-altering ultrasonic oscillations during the use of comparatively long ultrasound stimuli could possibly be the main effective factor for the induction of pain sensations and can also change the thresholds of other sensations (thermal, hearing and so forth).**

*Key Words:* **Focused ultrasound, Stimulation, Neural structures, Receptors, Sensations, Diagnosis, Mechanisms.**

## INTRODUCTION

The use of focused ultrasound in medicine and physiology for local destruction of deep-seated tissues in humans and animals is well-known and has been described elsewhere (Fry 1978; Fry et al. 1954; Gavrilov and Tsirulnikov 1980; Hill 1994; Wells 1977). Less is known about the possible application of focused ultrasound for inducing reversible changes of function in the activity of neural structures (Fry et al. 1950, 1958; Wall et al. 1953). Fry et al. (1950) have reported a temporary inhibition of the spontaneous activity in the excised crayfish ventral nerve cord. Temporary suppression of the functional activity in the central nervous system of animals was shown using focused ultrasound (Fry et al. 1958); however, the ultrasound doses required to obtain these effects were close to those that would destroy the tissues and would therefore have no practical use. Thus, all recorded effects were related to a "negative" stimulation, *i.e.,* with the suppression of the functional activity of neural structures. Focused ultrasound was also used more recently for the modification of electrically evoked responses from an *in vitro* hippocampal preparation (Rinaldi et al. 1991). It was shown that single pulses of focused ultrasound could induce a significant modification of nerve excitability *in vitro* for a period 40–50 ms following pulse termination (Mihran et al. 1990). In a theoretical work, Fry (1968) discussed the possibility of stimulating neural structures by the interaction of high-frequency and high-intensity electrical and ultrasound fields. Realization of this idea would require very powerful ultrasound and electrical fields which would be inappropriate because of possible hazards.

Research over a period of 20 years or so has demonstrated that focused ultrasound stimuli of short duration and relatively high intensity can induce a variety of somatic sensations, *e.g.,* tactile, thermal (warm and cold) and pain, without any attendant damage to tissues (Gavrilov 1984; Gavrilov et al. 1976a, 1976b, 1977a, 1977b; Gavrilov and Tsirulnikov 1980; Vartanyan et al. 1985). More recently, the ultrasonic method for the stimulation of neural structures was applied to research into joint pain in humans (Wright and Davies 1989; Wright et al. 1993). The other part of our research was related to the use of focused ultrasound for the stimulation of neural structures of the internal ear

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(Gavrilov 1984; Gavrilov et al. 1977c, 1980; Gavrilov and Tsirulnikov 1980; Tsirulnikov et al. 1988; Vartanyan et al. 1985). Auditory responses and phenomena produced by pulsed ultrasound in humans and animals have also been reported by other groups (Foster and Wiederhold 1978; Magee and Davies 1993).

The subject of the present review is restricted to the possibility of using focused ultrasound stimuli for nondestructive, long-term and repeated stimulation of superficial and deep-seated neural structures to induce different somatic, hearing and other sensations. Such possibilities are of interest for the application of focused ultrasound stimulation in physiological research and in clinical practice. The main advantages of using focused ultrasound are:

1. The technique is essentially noninvasive, *i.e.,* it does not require surgical intervention to stimulate deep-seated neural structures.
2. The locality of the stimulation can be controlled and changed by altering the resonant frequency of the ultrasound transducers to stimulate predetermined volumes of tissues and selected neural structures.
3. Precise control of stimulus parameters, *e.g.,* intensity, duration of stimuli, volume of stimulated region and so forth, is possible.
4. The possibility of inducing a wide variety of different sensations, both superficially and at different depths within tissues, is available with the same equipment.

Some of the results obtained in our three groups in this field are presented. The possible mechanisms of the stimulating effects of focused ultrasound and probable directions of future research are also discussed.

The studies reported were carried out after approval by the Research Ethical Committee at The Queen's University (Belfast, UK) and the Scientific Council of the Institute of Evolutionary Physiology and Biochemistry (Sankt-Petersburg, Russia).

## METHODS

### *Stimulation of receptors in skin and soft tissues*

Several modifications of the experimental methods were used in the investigations of somatosensory perception in humans. The focused ultrasound transducers with resonant frequencies from 0.48 to 3.0 MHz and the specially designed ultrasonic generators used in these studies, as well as the techniques applied for calibration of the ultrasound fields, have been described elsewhere (Gavrilov 1984; Gavrilov and Tsirulnikov 1980). For example, in Table 1, the calculated dimensions of the focal region of some focused transducers used in our early experiments are presented (Gavrilov et al. 1976a). In Table 1, $f$ is the resonant frequency of the focused transducer, $d$, $l$, $S$ and $V$ are the diameter, length, area and volume, respectively, of the focal region within the main diffraction maximum of its spatial distribution. Although the dimensions of this maximum, determined by points where the ultrasound intensity diminishes to zero, are relatively large (Table 1), the region where the intensity approximates to its peak value is considerably less: for the frequency of, *e.g.,* 2.67 MHz, the diameter of the focal region would be equal to fraction of a millimeter. Thus, using focused ultrasound transducers of relatively high resonant ultrasound frequency one can stimulate neural structures within relatively small volumes. The dimensions of focal regions of focused transducers, as well as the space distributions of their acoustic fields, were measured in water in free-field conditions by means of portable hydrophones (Gavrilov and Tsirulnikov 1980) and specially developed remote methods of measurements based on the use of focused ultrasound receivers (Gavrilov et al. 1988). Usually, such measurements gave good agreement between the theoretical and experimental results.

Table 1. Sizes of the focal regions of the focused transducers.

| $f$ (MHz) | $d$ (mm) | $l$ (mm) | $S$ (mm$^2$) | $V$ (mm$^3$) |
|---|---|---|---|---|
| 0.48 | 6.4 | 34 | 32 | 725 |
| 0.887 | 3.4 | 18 | 9.1 | 110 |
| 1.95 | 1.5 | 8 | 1.8 | 9.6 |
| 2.67 | 1.1 | 6 | 1.0 | 4.0 |

A schematic drawing of the installation for the study of the stimulation effects of focused ultrasound on the neural structures of the hand and forearm is presented in Fig. 1. The focused transducer was mounted on coordination equipment such that the three-dimensional position of the transducer could be controlled to within 0.1 mm. The hand and forearm of a human volunteer were constrained in a specially made mold of silumin (a light alloy of aluminum, silicon and some other elements) and submerged in a bath with water of variable but thermostatically controlled temperature, from 15° to 45°C, adjacent to the transducer (Gavrilov et al. 1976a, 1976b, 1977a; Gavrilov and Tsirulnikov 1980). The transducer focal length (usually 70 mm) permitted the stimulation not only of skin and superficial neural structures, but also of subcutaneous or deep-seated sites of the hand. Stimuli of ultrasound with frequencies of 0.48, 0.88 and 2.67 MHz and for durations of 1, 10 and 100 ms were generally used (Gavrilov et al. 1976a, 1976b, 1977a). In the preliminary experiments, the intensities of ultrasound, corresponding to definite values of the voltages at the focused transducers, were measured in free-field condi-

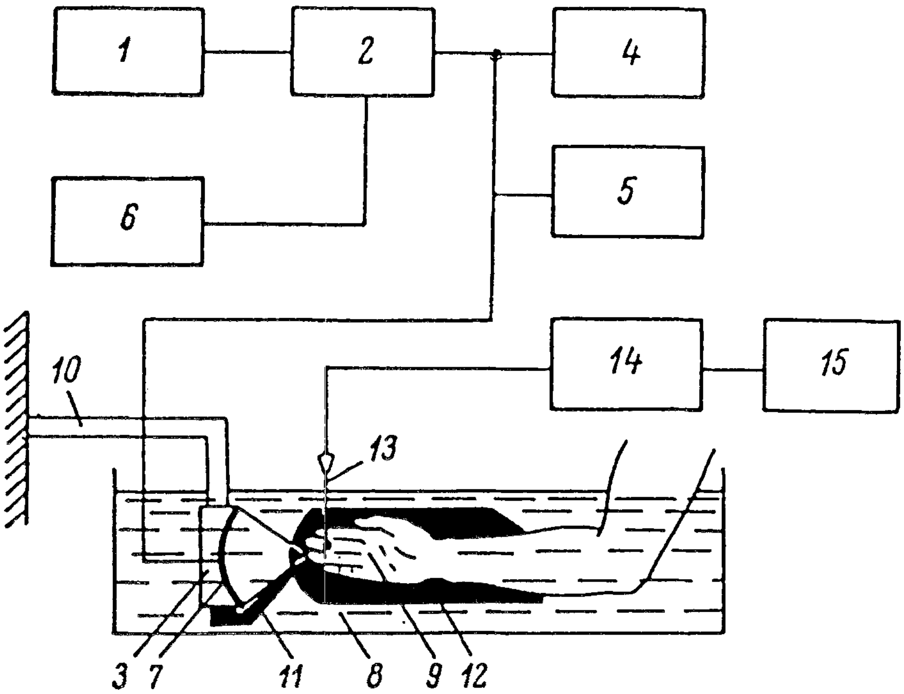


Fig. 1. A schematic drawing of the set-up for the study of the stimulation effects of focused ultrasound on the receptor neural structures of the hand and forearm (Gavrilov and Tsirulnikov 1980). 1—Ultrasound generator; 2—modulator and power amplifier; 3—housing of the focused transducer; 4—voltmeter; 5—oscilloscope; 6—generator of rectangular electrical pulses; 7—concave piezoceramic plate of the focused transducer; 8—bath with water of controlled temperature; 9—hand and forearm under investigation; 10—coordination equipment; 11—movable pointer of the center of the focal region; 12—mold of light alloy to keep a hand and arm in physiologically convenient position; 13—miniature hydrophone for cavitation measurements; 14—highly sensitive selective voltmeter-amplifier tuned to a frequency of one half the ultrasound frequency; 15—oscilloscope.

tions. Stimuli were applied, after a warning, at irregular intervals, the intensity increasing from stimulus to stimulus until a sensation was reported by the volunteer. The subject was asked to describe its character. The intensity of the focused ultrasound stimuli required to induce different sensations was recorded, from which other parameters, such as the displacement amplitude, the sound pressure amplitude, the temperature elevation and the radiation force and pressure could be calculated. The stimulated points were marked on the skin and these marks were redrawn onto the photograph or schematic drawing of the hand of the volunteer. For example, a schematic drawing with investigated points of one volunteer is presented in Fig. 2. Such "maps" enabled the same points to be stimulated on subsequent occasions.

A modified focused transducer with a truncated cone-shaped housing filled with water was used on other occasions. The hand or forearm was placed above the open end of the truncated cone and the acoustic contact between the piezoceramic plate of the transducer and the hand was accomplished through water. For some applications, a thin ultrasound-transparent film was attached to the open end of the truncated cone and the acoustic contact with the skin was provided by means of a thin coating of liquid paraffin.

*Induction of pain in joints*

A 500 kHz ultrasonic transducer was mounted on a stereotaxic targeting system immersed in a waterbath at 37°C and its position above the hand adjusted so that the focal region of the ultrasound beam was projected into the proximal interphalangeal joint. A variable-voltage 500 kHz input signal was amplified using a broadband power amplifier, the output of which was applied to the ultrasound transducer to produce stimuli of different intensities. The ultrasound transducer had a diameter of 36 mm and a focal length of a 40 mm. The stimulus duration of 100 ms and the variable interval between stimuli were controlled from a microcomputer which also recorded the evoked potential from the brain using the Data Acquisition in Neurophysiology (DAN) software package (Wright and Davies 1989; Wright et al. 1993).

*Stimulation of auditory structures*

Amplitude-modulated focused ultrasound was used to stimulate the neural structures of the labyrinth

| | | The ultrasound threshold intensity (W/cm²) | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | *Sensations at the skin* | | | | *Sensations under the skin* | | | |
| Spot No. | No. of replicate tests | Touch | Warmth | Cold | Pain | Touch | Warmth | Cold | Pain |
| | | | Finger | | | | | | |
| 1 | 1 | 20 | * | * | * | | | | |
| 2 | 1 | 44 | * | * | * | | | | |
| | 2 | 18 | * | * | * | | | | |
| 3 | 1 | 15 | * | * | * | | | | |
| | | | Palm | | | | | | |
| 4 | 1 | 75 | 385 | 980 | * | | | | |
| 5 | 1 | 80 | * | 880 | 900 | | | | |
| | 2 | 80 | * | * | 680 | | | | |
| 6 | 1 | 80 | * | 980 | 1180 | | | | |
| 7 | 1 | 27 | 680 | * | * | | | | |
| 8 | 1 | 60 | 980 | 980 | 1380 | | | | |
| | 2 | 55 | * | * | * | | | | |
| 9 | 1 | 44 | * | * | 880 | | | | |
| | 2 | 27 | 980 | * | * | | | | |
| | 3 | | | | | * | * | * | 520 |
| | 4 | | | | | * | * | * | 650 |
| | 5 | | | | | * | * | * | 520 |
| 10 | 1 | | | | | * | * | * | * |
| | 2 | | | | | * | * | * | * |
| | 3 | | | | | * | * | * | * |
| | 4 | | | | | * | * | * | * |
| 11 | 1 | | | | | * | * | * | * |
| | 2 | | | | | * | * | * | * |
| | | | Fore arm | | | | | | |
| 12 | 1 | 85 | * | 1100 | * | | | | |

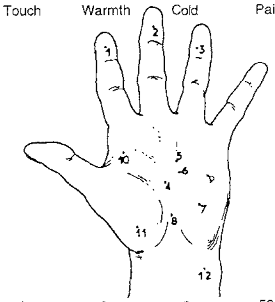


Fig. 2. A schematic drawing of the hand of one of the volunteers with spots studied at the frequency 0.887 MHz and pulse duration 1 ms. The table gives the ultrasound intensities corresponding to different threshold sensations in the various locations. Asterisks denote that the sensation of a given modality was absent (Gavrilov et al. 1976a).

of the ear (Gavrilov 1984; Gavrilov et al. 1977c, 1980; Gavrilov and Tsirulnikov 1980; Tsirulnikov et al. 1988; Vartanyan et al. 1985). Figure 3 shows a part of the experimental setup for the stimulation of the neural structures of the auditory system. The precise location of the focused transducer was achieved by a special coordination device which allowed movement of the transducer to be controlled in three mutually perpendicular directions with an accuracy of 0.5 mm. The transducer (Fig. 3) was placed in a sound-transparent polyethylene bag filled with water heated to 30°–35°C. A removable focus pointer (Fig. 3) was mounted to the transducer to locate the centre of the focal region. The subject's head was placed on a pillow so that the acoustic axis of the transducer was perpendicular to the skin in front of the external ear. The focus pointer was then removed and the polyethylene bag was lowered until it was in acoustic contact with the skin. For better contact the skin was coated with liquid paraffin. Thereafter, the transducer was moved 30–35 mm closer to the skin by means of the coordination device. It was important to bring the direction of the focused radiator's acoustic axis into coincidence with the labyrinth of the ear. Under these conditions, the intensities of the stimuli that produced the thresholds of hearing sensations were at a minimum. A more adaptable version of the equipment for the use of focused ultrasound in clinical research has also been developed (Tsirulnikov et al. 1988b).

Ultrasound was amplitude-modulated by sine-shaped signals at standard audiometric octave frequencies in the range of 125–8000 Hz. Pulse modulation was also used with pulse duration from 0.05–0.1 ms with the same repetition frequencies. Pulse-amplitude modulated ultrasound was also used in several studies.

Hearing thresholds were expressed in ultrasound intensity values. In the research on hearing loss in patients, these thresholds were compared with the thresholds in people with normal hearing. Levels corresponding to degrees of hearing loss were marked on standard audiograms for air and bone conduction. A comparison was made of tonal and ultrasonic audiograms from patients to reveal some typical signs that could be of use for diagnosis of various hearing disorders.

## STIMULATION EFFECTS OF FOCUSED ULTRASOUND IN PHYSIOLOGICAL RESEARCH

The physical and technical background of the use of focused ultrasound in medicine and physiology has been described in a number of studies (Fry 1978; Gavrilov and Tsirulnikov 1980; Hill 1994; Kossoff 1979; NCRP 1983; Nyborg 1985; Sanghvi 1994; Wells 1977). Here it is appropriate to note only that the dimensions of the focal regions of the focused transducers depend mainly on the ultrasonic frequency and the angle of convergence of the ultrasonic beams. For example, for a typical transducer used in these studies with a resonance frequency of 1 MHz, a radius and focal length of 42 and 70 mm and an angle of convergence of 36°, the diameter and the length of the focal region would be 3 and 15 mm, respectively.

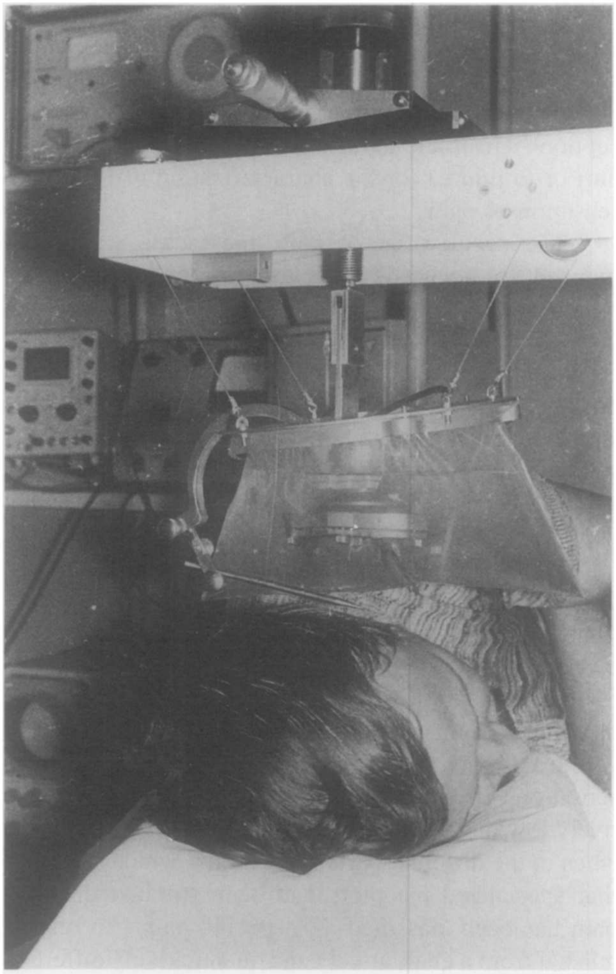

Fig. 3. A part of the experimental set-up for the stimulation of the neural structures of the auditory system (Gavrilov 1984; Vartanyan et al. 1985).

Using short and rather intense pulses of focused ultrasound directed into sensitive points in a human hand and arm or in other parts of the body, it was possible to produce many somatic sensations, the character of which depended on the parameters of focused ultrasound and the location of the focal region (Gavrilov et al. 1976a, 1977a). By changing the intensity and durations of the stimuli and also the location of the focal region, all sensations that humans can perceive through their skin, *i.e.*, tactile, temperature (hot and cold), different pain sensations and so forth, were induced. Pain, as well as some kinds of tactile sensations, can be induced by focused ultrasound not only in the skin neural structures, but also in the deep-seated tissues. The results of one set of such studies in one volunteer with typical values of threshold intensities of ultrasound ($f = 0.887$ MHz) corresponding to different somatic sensations in various investigated spots, are presented in Fig. 2 (Gavrilov et al. 1976a).

The threshold values of the ultrasound intensities corresponding to the induction of different sensations with the use of different ultrasound frequencies and stimulus duration of 1 ms are presented in Table 2 (Gavrilov et al. 1976a). These data were obtained from the lowest intensities observed at the same points in two volunteers. The number of spots investigated in every location varied from 5 to 21, and the number of measurements in every spot was varied from 4 to 42 (see details in Gavrilov et al. 1976a).

The use of focused ultrasound as a new convenient stimulus yielded data which were of interest in the study of sensory perception *per se* (Tsirulnikov 1993; Vartanyan et al. 1985). For example, the existence of "universal" temperature-sensitive points which could be related to warm and cold sensations was shown, in contrast to the widespread conception that there are separate warm- and cold-sensitive points. The results of investigations into a large number of different points and in different parts of the body suggested that temperature-sensitive points can function as warm or cold ones depending on the ambient temperature (Gavrilov et al. 1976a, 1977a).

During investigations into the intensity of the focused ultrasound required to achieve the minimum threshold for a tactile response on the arms of healthy volunteers, it was found that these thresholds increased in the direction from fingers to forearm (Tsirulnikov et al. 1982). Thresholds at responsive points on the forearm were always higher than those on the fingers and palms. All these data suggest that a value of the threshold could depend on the density distribution of the receptors (Tsirulnikov et al. 1982).

Special attention was paid to research into pain induced by a stimulus of focused ultrasound (Gavrilov et al. 1976a, 1977a; Tsirulnikov and Gurgenidze 1990; Tsirulnikov et al. 1985, 1986; Wright and Davies 1989;

Table 2. The threshold values of the ultrasound intensities. (W/cm$^2$) corresponding to the induction of different sensations with the use of stimulus duration of 1 ms and different ultrasound frequencies (Gavrilov et al. 1967a).

| | | Frequency (MHz) | | | |
|---|---|---|---|---|---|
| Sensations | Localisation | 0.48 | 0.887 | 1.95 | 2.67 |
| Tactile | Finger | 8 | 15 | 80 | 120 |
| | Palm | 16 | 80 | 250 | 350 |
| Warm | Finger | 55 | 90 | 1420 | 3200 |
| | Palm | 130 | 820 | 2940 | 4500 |
| Cold | Finger | — | — | — | — |
| | Palm | 130 | 820 | 2000 | 3000 |
| Pain | Finger | 55 | 140 | 2860 | — |
| | Palm | 290 | 350 | — | 3000 |

—: Absence of sensation with maximum intensity of stimulus.

Wright et al. 1993). It was found that, depending on the location of the centre of the focal region, pain sensations, namely pain in skin, in soft tissues, in bone and in the joint, differed markedly in the intensity required to achieve thresholds, the area of the pain irradiation and in subjective characteristics of the response (Gavrilov et al. 1976a, 1977a). Threshold intensities for all sensations, including pain, decreased markedly when the duration of the stimulus was increased from 50 to 500 ms (Vartanyan et al. 1990).

A separate series of experiments was carried out to evaluate the possible influence of ultrasonically induced cavitation in soft tissues on the origin of pain sensations (Gavrilov and Tsirulnikov 1980). Cavitation was controlled by recording the subharmonic components of the acoustic noise arising when cavitation appeared in the medium. The details of such measurements in brain tissues *in vivo* in animals have been published (Gavrilov 1974). An experimental set-up included a specially designed miniature hydrophone placed in water within 20–30 mm of the centre of the focal region, a selective voltmeter-amplifier tuned to a frequency of one half the ultrasound frequency and an oscilloscope to record the subharmonic acoustic noise (Fig. 1). The intensity of ultrasound, for which the amplitude of the acoustic signal on the screen of the oscilloscope exceed by 40 dB the level of the initial acoustic noise, was considered the cavitation threshold (Gavrilov and Tsirulnikov 1980).

With a stimulus duration of 1 ms, no pain sensation, as a rule, was reported, although violent cavitation was registered. With a stimulus duration of 10 ms, the threshold intensities for pain exceeded the intensities corresponding to cavitation thresholds. At a stimulus duration of 100 ms, cavitation appeared at lower intensities than pain in some cases but, in other cases, when pain was experienced, cavitation was absent. Naturally, in these particular experiments, it was rather difficult to separate definite cavitation occurring in the tissues from that in water; however, in terms of possible correlation between pain and cavitation, the most noteworthy cases were those in which pain was experienced but cavitation was not registered. Thus, in these experiments, there was no defined causal relationship revealed between the appearance of cavitation and threshold sensations of pain.

Naturally, such experiments could not give a direct and unambiguous explanation of the relationship between the ablation of the tissue and the induction of pain. From this point of view, another of our studies was more informative, in which it was found that pain in a defined sensitive point in one volunteer (one of the present investigators) could be reproduced many times over a number of years without a marked change in the pain threshold. Thus, at least in our experiments, threshold pain was not associated with the destruction of, or injury to, tissues. This important conclusion suggests that it is not always correct to use the term ''nociception'' (from the Latin word ''nocivus'' meaning to hurt or to injure) for the characterization of threshold sensation of pain.

The peculiarities of pain and tactile sensations induced by focused ultrasound in the acupuncture points and in arbitrary deep-seated sensitive points were investigated (Tsirulnikov et al. 1986; Vartanyan et al. 1985). It was presumed that in the acupuncture points there was a greater preponderance of receptor structures related with pain. The sensations after ultrasonic stimulation of the acupuncture points were similar to so-called ''foreseen'' sensations induced by the use of needles for acupuncture.

A detailed investigation of the subjective characteristics and the threshold intensity for skin pain sensations in the different parts of the human body was carried out (Tsirulnikov and Gurgenidze 1990). Maps of the distribution of the thresholds of specific skin pain in the human body were drawn. In the majority (up to 90%) of the 3750 points investigated, the pain sensations in superficial, cutaneous layers had relatively low thresholds and similar qualitative characteristics in the different parts of the body, which suggests that specialized receptors had been stimulated. Such pain has been classified as ''specific pain'' to distinguish it from a qualitatively and quantitatively different pain sensation, which has been classified as ''nonspecific'' pain. Subjectively, this ''nonspecific'' pain was far more ''unpleasant'' and frequently associated with a ''burning'' sensation. The intensities of focused ultrasound required to induce ''nonspecific'' pain differed markedly in various parts of the body and the pain probably resulted from a superintense stimulation of tactile or temperature receptors. The threshold intensity for ''nonspecific'' pain increased from the fingers to the elbow (Tsirulnikov et al. 1985).

Pain produced by projecting a focused ultrasound stimulus at 0.5 MHz for 100 ms into the interphalangeal joint of the hand in human volunteers was monitored by recording the brain-evoked potential. This recording was obtained by the summation of 80 stimuli, 40 of which were at an intensity of 1.5 times that of the threshold of pain perception and 40 at twice the threshold intensity (Wright and Davies 1989). The pattern of the evoked potential produced as a result of the stimulation of the nociceptors in the joint was identical to that obtained from a superficial, painful electrical stimulation from electrodes placed on the skin of the finger. In both cases, three peaks of electrical activity in the brain were recorded within 1 s from the initiation of the stimulus—a small, first positive peak (P1) followed by a first negative peak (N1), both of which were sharp, and finally a broad second positive peak (P2). The change in potential between N1 and P2 provided a measure of the amplitude of the evoked potential which showed no statistically significant change over a period of 4.5 h. A subjective assessment of the painfulness of the stimuli, measured concurrently using a visual analogue scale score, showed a significant correlation with the evoked potential amplitude, and the latter could therefore be used as an objective measure of the perceived pain and provide a useful model for the study of joint pain (Wright and Davies 1989).

A separate, large field of investigations was related to the use of focused ultrasound for the introduction of auditory information in humans (Gavrilov 1984; Gavrilov and Tsirulnikov 1980; Gavrilov et al. 1977c, 1980; Tsirulnikov et al. 1988b; Vartanyan et al. 1985). In the initial experiments on frogs it was shown (Gavrilov et al. 1977b) that, under the action of stimuli of focused ultrasound on the labyrinth structures, the electrical responses recorded in the midbrain auditory area were similar to responses to sonic stimuli

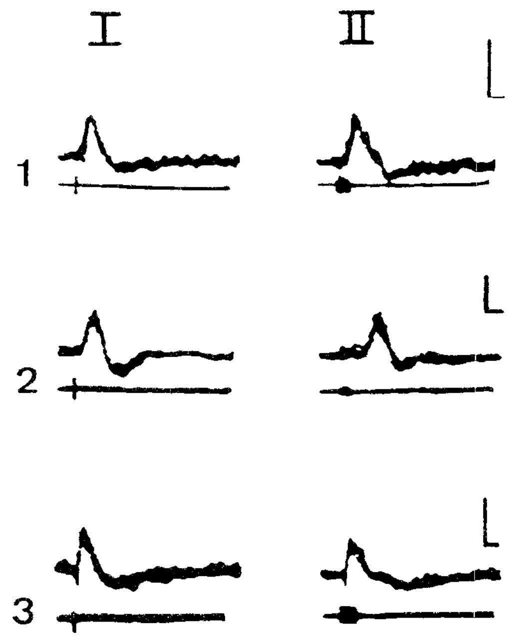


Fig. 4. Evoked responses in the midbrain auditory area of the frog. I: Stimulation of the labyrinth with focused ultrasound (frequency 0.48 MHz; stimulus duration 1 ms); II: response to sonic stimuli of frequency 100–1600 Hz (20 ms duration). Stimulus intensity: 20 dB above threshold. Top: the oscillogram of responses; bottom: the stimulus. Calibration (at right side of recordings): vertical axis 100 $\mu$V, horizontal axis 25 ms. Nos. 1–3: the animal's number (Gavrilov et al. 1977b).

(Fig. 4). The ultrasound intensities applied in these experiments were 100–1000 times lower than intensities that cause structural changes in the tissues (Fry 1978; Gavrilov and Tsirulnikov 1980).

As a result of further investigations, a new method for the introduction of auditory information in humans has been suggested (Gavrilov et al. 1980; Gavrilov and Tsirulnikov 1980; Tsirulnikov et al. 1988b). The method is based on the stimulation of neural structures of the labyrinth of the ear by focused ultrasound which is amplitude-modulated by an information signal at audio-frequency. In this case, the auditory information introduced to a human labyrinth corresponded to the envelope of the amplitude-modulated ultrasonic signal.

Focused ultrasound was able to stimulate, through the skull, not only the receptors (hair cells) of the internal ear, as with ordinary sound stimulation, but also the auditory nerve fibers. This result was supported by experiments on intact frogs and on frogs in which the receptors in the labyrinth had been destroyed. In the latter case, the responses to the ultrasonic stimulation in the midbrain auditory centres were recorded, but at much higher threshold intensities (Vartanyan et al. 1981). Histochemical methods confirmed that, in the case of receptor system disorders, ultrasound did activate the nerve fibers (Vartanyan et al. 1981). Finally, the possibility of direct stimulation of auditory nerve fibers by focused ultrasound is supported by the fact that some completely deaf persons, whose receptor systems were diagnosed to have been destroyed, may perceive auditory information delivered by means of amplitude-modulated focused ultrasound, whereas ordinary hearing aids were not effective (Gavrilov and Tsirulnikov 1980). The proposed method may be useful to reveal those patients whose hearing function might be successfully improved by electrical cochlear implants.

Focused ultrasound could provide a universal means for the stimulation of different neural structures. For example, ultrasound stimuli directed into the tongue tissues can induce very singular taste sensation. Stimulation of the electroreceptors in the skate induced an increase in the spike activity (Broun et al. 1980). Experiments on invertebrates (edible snails without shells) showed the possibility of using focused ultrasound to stimulate not only the receptor structures, but also the central nervous structures (Gavrilov et al. 1978). The results of these studies form the basis of similar investigations on vertebrates including mammals.

## STIMULATING EFFECTS OF FOCUSED ULTRASOUND IN CLINICAL PRACTICE

There are grounds to suppose that ultrasonic methods for stimulating neural structures should find wide and useful application in the diagnosis of various diseases related to a change of sensory perception from normal. For example, a number of neurological and skin diseases are accompanied by considerable differences in the sensitivity of skin or tissue receptors from normal. Thus, by measuring and comparing the thresholds of various sensations in normal and pathological states, one may prove the diagnosis, evaluate the extent of pathological damage, monitor the results of treatment and so forth. Such investigations have been carried out by measuring tactile sensitivity in 30 patients with various neurological diseases, *e.g.*, syringomyelia, radiculitis spondilogena cervicalis, cerebral apoplexy and others (Gavrilov and Tsirulnikov 1980; Godovanik et al. 1978). All patients exhibited a deviation from normal tactile sensitivity, ranging from a considerable increase of tactile thresholds up to entire absence of the tactile sensitivity. Changes of tactile thresholds were revealed not only on the affected side of the body but also on the opposite, intact side. This method provides not only the means for detecting or confirming sensitivity disorders but also for characterizing them quantitatively, evaluating the extent of the disorders for the various forms of pathology and revealing and identifying "subclinical" neurological disorders, which traditional diagnostic methods fail to reveal.

Focused ultrasound has also been used to measure sensitivity thresholds in the skin of the hand in 12 normal persons and 42 patients with dermatoses, *e.g.*, psoriasis, neurodermitis, sclerodermia, lymphoma cutis and others (Tsirulnikov et al. 1988). Thresholds in patients were measured only in the visually unchanged, unaffected parts of the skin. It was found that tactile thresholds in all patients were higher than in healthy persons.

The comparative study of the pain thresholds in 51 healthy persons and 64 patients with neurasthenia, which is a widespread kind of neurosis, was carried out (Ashkinazi et al. 1992). This disease is a natural model of chronic psychoemotional stress. These patients show a distinct tendency to a decrease of pain thresholds, a lowered adaptation to repeated stimuli and a sensitization to pain perception.

In research into healthy children as well as into children with inborn and postamputation stumps of the forearm, the relationship between the level of tactile sensitivity in the skin of the forearm and the level of its muscular motor activity has been studied (Tsirulnikov et al. 1990). These studies were performed using focused ultrasound stimulation in children of different ages. It was found that, with the increase in muscle activity of the forearm, irrespective of age, tactile thresholds decrease.

The joint pain model just described was used in an assessment of the analgesic action of pethidine. The progressive diminution in the amplitude of the evoked potential over a period of 3.5 h following the administration of the drug showed a significant correlation with the corresponding visual analogue scale score (Wright et al. 1993). In view of this correlation, and since the intensity of the stimuli over the period of the trial was not altered, it was suggested that the evoked potential provided a reliable, objective measure of the perception of pain (Wright et al. 1993). Restoration of the amplitude of the evoked potential to its original, predrug value by increasing one or more of the parameters of the focused ultrasound stimulus would suggest that the value of those parameters that were required to achieve the threshold of pain had also increased. The measurement of those parameters in a clinical setting could provide a quantitative measure of the sensitivity of diseased or damaged joints and could be used to follow the progress of ameliorative therapy.

The characteristics of joint pain induced by focused ultrasound were evaluated qualitatively using the McGill pain questionnaire and compared with the pain experienced by patients with arthritis of the hand joints. The sensory qualities of focused ultrasound-induced pain and the pain experienced by arthritic patients were found to be similar, more so in the case of osteoarthritis than rheumatoid arthritis (Wright 1989). This similarity in the sensory characteristics of the pain in each case makes the focused ultrasound-induced joint pain model a clinically useful correlate for the evaluation of different therapeutic treatments of arthritic pain.

The proposed method of introduction of auditory information to humans, based on the use of amplitude-modulated focused ultrasound, was tested at the Research Institute on Ear, Throat, Nose and Speech (Sankt-Petersburg) for the diagnosis of various hearing disorders (Tsirulnikov et al. 1988). Measurements of hearing sensation thresholds as a result of the stimulation of the neural structures in the labyrinth of the ear by amplitude-modulated focused ultrasound were carried out. Ultrasound oscillations were modulated by sinusoidal signals at standard audiometric octave frequencies. Then, the ultrasonic frequency–threshold curves, so-called "ultrasonic audiograms," were compared with ordinary audiograms, *i.e.*, audiometric curves for bone and air conduction. Evidently, the propagation paths of the sonic signal to the labyrinth in cases of the usual auditory stimulation and amplitude-modulated ultrasound stimulation may be essentially different. Consequently, the frequency–threshold curves obtained by these two methods should also vary. The distinctive features of ultrasonic audiograms and the difference between these curves and the traditional audiometric ones were established and used as diagnostic criteria (Tsirulnikov et al. 1988). To date, more than 5000 patients have been investigated. The ultrasonic method can be used successfully for diagnosis, especially in complicated clinical cases with compound pathology, when traditional audiometric methods do not provide an unambiguous, reliable diagnosis. A number of patients were investigated specifically for the evaluation of the diagnostic effectiveness of the developed method. Among these patients, 488 had chronic cochleaneural hearing loss, 122 had acute cochleaneural hearing loss, 230 had otosclerosis and 70 had Meniere's disease. For patients with cochleaneural hearing loss, the accuracy of diagnostic and prognostic tests approached the absolute level (near 100%), for otosclerosis 98% and for Meniere's disease 76%. If the ultrasonic tests were combined with complex clinical investigations, the trustworthiness of the diagnosis approached to 100%; that is, the diagnosis was highly reliable.

Thus, the preliminary results of the use of focused ultrasound in clinical practice for the stimulation of neural structures are highly promising and hopeful.

## POSSIBLE PHYSICAL MECHANISMS OF STIMULATION EFFECTS OF FOCUSED ULTRASOUND

It is evident that a better understanding of the mechanisms underlying the stimulating effects of focused ultrasound is very important for effective and safe application in physiological research and in medical practice. Here we present some considerations related to the possible physical mechanisms of the stimulation of receptor neural structures by focused ultrasound.

In attempts to reveal the main factor responsible for the stimulating effects of focused ultrasound, the thresholds of different sensations in the same sensitive points using different ultrasound frequencies, *i.e.*, from 0.48 to 2.67 MHz, have been analyzed. In other words, we were trying to investigate the frequency dependence of stimulation effects of focused ultrasound at receptor structures. The purpose of this analysis was to reveal the parameter of focused ultrasound that remained unchanged at the thresholds for the stimulation of the same points using focused ultrasound of different frequencies. Using the threshold intensities of focused ultrasound, which we have measured for every kind of sensation, we have calculated the values of all the possible parameters of focused ultrasound (sound pressure, power and energy of pulses, elevation of temperature in the tissue, particle velocity, acceleration, radiation pressure, radiation force and so forth) that could be potentially responsible for the stimulating effects of focused ultrasound.

These calculations are simplified by the fact that, through the main diffraction maximum of the focal region, the ultrasound beam is propagated as a plane wave. Therefore, for simplicity, these calculations were fulfilled by the use of the relationships applicable to plane progressive monochromatic ultrasound waves (Bergmann 1954):

$$I = \frac{1}{2}\rho c \omega^2 A^2 = \frac{1}{2}\rho c V^2 = \frac{P^2}{2\rho c} = \frac{W}{S} \qquad (1)$$

where $I$ is the time-averaged intensity of ultrasound; $A$ is the amplitude of displacement; $V$ is the amplitude of the oscillatory velocity; $P$ is the amplitude of the sound pressure; $W$ is the acoustic power of the transducer; $\omega = 2\pi f$ is the angular frequency ($f$ is the ultrasound frequency); $\rho c$ is the characteristic acoustic impedance of the medium of density $\rho$, and in which the velocity of sound is $c$; and $S$ is the area of the focal region of the transducer, which is inversely proportional to $f^2$. In eqn (1), the quantities $I$, $A$, $V$ and $P$ are independent of position. In reality, most ultrasonic fields are spatially nonuniform, but the above relationships are nevertheless locally applicable in most cases (NCRP 1983).

The values of temperature elevation, $\Delta T$, in the tissue at the end of the ultrasound pulse were calculated from the following equation derived with allowance for the thermal conductivity of the tissue (Pond 1970):

$$\Delta T = 2\alpha I t A(t)/J\rho C_p \qquad (2)$$

where $\alpha$ is the amplitude absorption coefficient, which is dependent on the ultrasound frequency $f$, $I$ is the intensity of ultrasound in the focal region; $t$ is the ultrasound pulse duration; $A(t)$ is a function taking into account the thermal conductivity of the tissue (Pond 1970); $J$ is the mechanical equivalent of heat; and $C_p$ is the specific heat of the tissue. For the calculations, the coefficient $\alpha$ in the soft tissues was assumed to be equal to 0.1 Np/cm MHz (NCRP 1983). For durations of ultrasound stimulus shorter or equal to about 100 ms it is possible to neglect by the effect of thermal conductivity in the tissues and consider $A(t) = 1$. But, for the longer stimulus durations, this effect should be taken into account and the value of $A(t)$ might well be calculated according to work of Pond (1970).

From the comparative analysis of all these parameters for different ultrasound frequencies (from 0.48 to 2.67 MHz), it was apparent that only one parameter, *i.e.*, the amplitude of displacement $A$ (which is proportional to the square root of the acoustic power), remained relatively constant for the different ultrasound frequencies. The other parameters of focused ultrasound changed markedly, sometimes over the range of several orders of magnitude.

This tendency is most clearly revealed for tactile sensations, since the values of these thresholds were always the most reproducible. Figure 5 illustrates the relative changes of some parameters over the frequency range from 0.48 to 2.67 MHz for tactile and thermal sensations at the same points of the human fingers (Gavrilov et al. 1976a; Gavrilov 1984). The duration of stimulus was 1 ms in each set of experi-

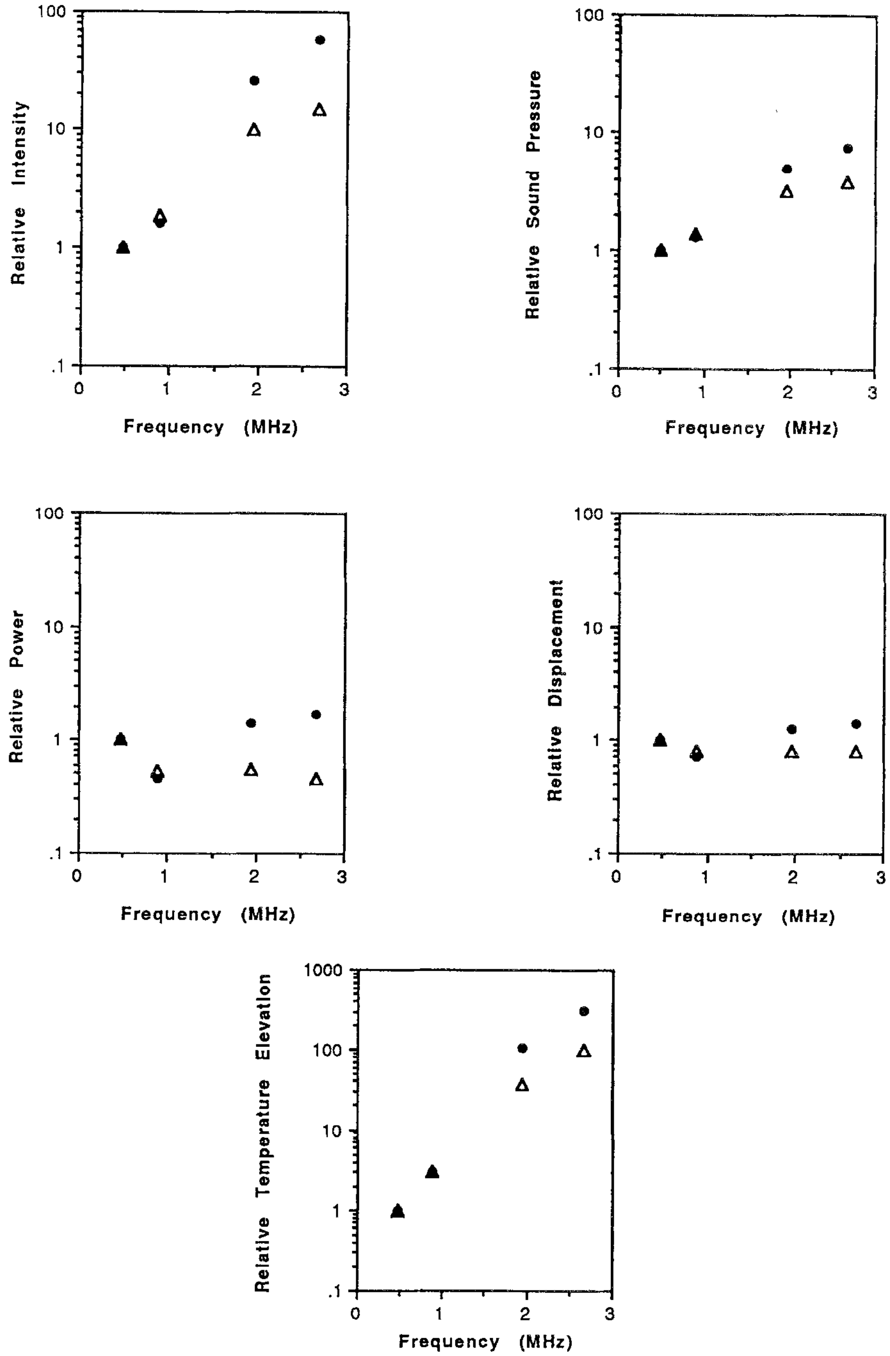


Fig. 5. Relative changes in the parameters of focused ultrasound at the frequency range of 0.48–2.67 MHz for tactile and thermal sensations in the hand induced by ultrasound stimuli. Abscissa: ultrasound frequency; ordinate: relative changes in parameters (the value of each parameter at a frequency of 0.48 MHz was taken as a unit). The duration of stimulus was 1 ms. (△) Tactile sensations; (●) thermal sensations (Gavrilov 1984; Gavrilov et al. 1976a).

ments. The values of each parameter were calculated relative to that at 0.48 MHz. From Fig. 5, it is apparent that the amplitude of displacement for tactile and thermal sensations remained relatively constant for different frequencies. In contrast, the power was more variable, the sound pressure was changed approximately sixfold, the intensity more than 30-fold and the elevation of temperature about 100-fold. This is clear because the power (and consequently the radiation force) is proportional to $A^2$, the sound pressure to $fA$, the intensity (and therefore the radiation pressure) to $f^2A^2$, as seen from eqn (1), and the elevation of temperature to $f^3A^2$, since it is proportional to $If$.

However, the direct correlation between the thresholds of tactile and thermal sensations and the values of the amplitude of displacement is not quite clear. In fact, the amplitude of displacement characterizes the oscillating, sign-altering displacement of the medium in the acoustic field. However, the actual mechanism of the stimulation of neural structures under the action of focused ultrasound should be related to a certain "unidirectional," "rectified" action rather than to a sign-altering oscillation displacement of the medium *per se*. For example, volunteers could not differentiate between tactile sensations in response to a long stimulus or two short stimuli within the same time interval, *i.e.*, a stimulus lasting 400 ms or two stimuli of 10 ms with an interval between them of 380 ms. The stimulus with the duration of 400 ms induced sensations only at the onset and termination of the stimulus.

At first sight, the radiation pressure or radiation force of ultrasound might be the most suitable candidates to provide such a "unidirectional" factor, due to their well-known physical nature (Bergmann 1954; Wells 1977). However, the radiation pressure can be excluded from consideration because it is proportional to the intensity of ultrasound and therefore changes very greatly with the change of the frequency (see, *e.g.*, Fig. 5). The radiation force, which is defined by the acoustic power of the transducer and, in general, is independent of the frequency, may be considered as the major potentially effective stimulating factor. In fact, an ultrasound beam with a power of 1 W traveling in water produces a force of $6.7 \times 10^{-4}$ N when it is completely absorbed (Wells 1977). This is equal to the force of gravity acting on a weight of about 69 mg. If the target is a perfect reflector, this force is doubled. Thus the action of the radiation pressure as well as the action of any other mechanical factor of focused ultrasound could explain its stimulating effects at mechanoreceptors.

The mechanical action of ultrasound may be also the main effective factor for the stimulation of thermal sensations (Gavrilov and Tsirulnikov 1980). There are data in the literature on the dependence of the contractility of skin's collagen with temperature. Therefore, one can propose that a change of temperature could turn into mechanical displacement due to the collageneous structures surrounding the receptive neural structure (Gavrilov and Tsirulnikov 1980).

From the results presented and others, it was concluded that, at least for the duration of ultrasound stimuli up to 100 ms, the main effective factor of focused ultrasound as a physiological stimulus for the induction of tactile and thermal sensations was neither "energetic" nor "thermal" in nature, but a mechanical one, similar to the action of a "push" at the receptor structures within the sensitive region. For stimuli of about 500 ms duration or more, the temperature elevation in the tissue may be important, being increased with the increasing stimulus duration. The mechanical effect of focused ultrasound at the plasma membrane of cells could result in changes in their permeability, especially to ions, that would cause a change in the membrane potential. Such mechanical effects have been reported in cells subjected to shear stress (Lansman et al. 1987; Olesen et al. 1988).

The situation with regard to pain sensations would appear to be more complex. Indeed, the action of the mechanical displacement at the beginning and the end of the stimulus may not be involved, since ultrasound stimuli of comparatively long duration but low intensity will induce a pain sensation which probably results from the release of biochemical mediators (*e.g.*, histamine) from adjacent cells that stimulate the nociceptors (Terenius 1981; Vartanyan et al. 1990). Under these circumstances, a radiation force of several grams would not induce pain, but when the pain threshold is exceeded the main effective factor of ultrasound is probably due to the sign-alternating oscillations of ultrasonic frequency.

On the other hand, it is apparent that, for short ultrasound stimuli, the origins of pain sensations are not related to the thermal action of ultrasound. In fact, it was possible to induce, at the same point, the same pain sensation using such parameters of focused ultrasound that resulted in the elevation in temperature in the tissue from 0.01°C to several tens of degrees, depending on the duration and intensities of the stimuli (Gavrilov and Tsirulnikov 1980; Gavrilov et al. 1976a). The mechanical action of focused ultrasound remains as the most probable effective factor for the induction of pain sensations only for very short and very intense ultrasound stimuli, like during the use of lithotripters.

The mechanism of the stimulating effects of amplitude-modulated ultrasound at the neural structures of the labyrinth was also studied (Gavrilov and Tsirulnikov 1980; Tsirulnikov 1993; Tsirulnikov et al. 1988; Vartanyan et al. 1985). First, it is necessary to keep in the mind that the hearing organ is an extremely sensitive device designed to perceive an action of adequate (*i.e.,* sound) information. Therefore, it is necessary to take into account the effect on the labyrinth of the sound component arising as a result of the radiation pressure of amplitude-modulated ultrasound. It is known that the radiation pressure of amplitude-modulated ultrasound propagating through the medium consists of three components: one constant and two variable. One of the variable components changes with the modulation frequency, *i.e.,* with the sonic frequency, and the other with double the modulation frequency (Altenberg and Kästner 1952; Bergmann 1954).

Thus, if the receptor system of the labyrinth in humans functions normally, the most probable factor inducing the auditory sensations in the hearing organ is the effect of sonic oscillations arising due to the variable component of radiation pressure on the receptors in the labyrinth. In this case, only sonic information is going to the labyrinth, whereas ultrasound serves as a means of its delivery. At the same time, ultrasound oscillations under defined conditions can change thresholds of tactile and hearing sensations (Tsirulnikov 1993; Vartanyan et al. 1981), increasing the sensitivity of the perceptive neural structures.

As previously mentioned, amplitude-modulated ultrasound may, under certain circumstances, induce a direct stimulating effect on the fibers of the auditory nerve (Vartanyan et al. 1981). The basic stimulating factor in this case still remains unclear, but could most probably be the mechanical one previously discussed.

Thus, depending on the parameters and regimes of the use of focused ultrasound (intensity, ultrasound frequency, duration and repetition frequency of stimuli, character of modulation and so forth) the origin of any functional effects lies with different characteristics of ultrasound or their combination. Thus, by selecting an appropriate regime of ultrasonic irradiation it is possible to stimulate differently various receptor structures.

## CONCLUSIONS

The results of the investigations carried out showed that focused ultrasound is a useful and promising means for the stimulation of both superficial and deep neural receptor structures. In fact, an accurate, quantitative measurement of the parameters of a stimulus that produces tactile, thermal (both warm and cold), pain and hearing sensations at precisely located, small, discrete regions both superficially (*e.g.,* in skin) and deep-seated within tissues (*e.g.,* in joints and muscles or in the labyrinth of the ear) provides an invaluable and universal means of assessing neuronal functions in humans. This method reveals differences in the neuronal structures and their sensitivity, which are involved in a specific response to the focused ultrasound stimulus. Changes in the parameters of stimuli required to elicit a selected response could provide valuable information of neuronal function as an aid in both the diagnosis of neuronal defects and in monitoring the effectiveness of therapy.

Although there is a variety of proposed methods and approaches, all have common basic features. They are all noninvasive, local, remote and universal. To date, two wide and prospective fields of medical applications of focused ultrasound for the stimulation of neural structures have arisen. The first is concerned with the stimulation of the receptor structures of the somatic system of humans. It is possible that, in the near future, a new broad field of ultrasonic medical diagnostics will open up, based on precise measurements and comparison of thresholds of various sensations in persons with normal perception and in patients with different kinds of pathology. This method has been used for diagnosis of neurological, skin and other diseases that involve changes of perception for different sensations. This technology can also be used to evaluate the success of prostheses for rehabilitation of partly or completely lost functions. The second large and promising field of medical application of the methods is related to diagnosis of hearing disorders and prosthesis of hearing function in some people with deafness.

Some prospective directions for future research in the field of clinical application include:

1. Application to other kinds of pathology, for which the use of the proposed methods of diagnosis, treatment and prognosis might prove useful.
2. Use of focused ultrasound for the relief of pain and for therapy for various diseases by means of local stimulation with focused ultrasound of predetermined deep structures, and particularly of acupuncture points.
3. Evaluation of the efficiency of anesthetic and analgesic drugs by measuring of pain thresholds before and after drug administration.
4. Assessment of the state of regeneration of the bone tissue after fractures by means of measurement of the pain thresholds at the periosteum.
5. Use of these methods and devices in prostheses, for the development of the apparatus for rehabilitation; *e.g.,* for optimal selection of individual prosthesis in which the feedback is based on tactile sensitivity, for the development of individual training pro-

grams, for prosthesis adaptation and for working out recommendations and prognoses of the results of individual prostheses.

6. Development of devices, technical means and equipment for the diagnosis and treatment of hearing disorders and for prostheses for hearing function in people with deafness.
7. Treatment of some diseases (in particular, hearing disorders) by the use of focused ultrasound with special drugs (some modification of phonophoresis).

The ultrasonic stimulation of neural structures is also of great importance in physiological research. Some possibilities for future research in this field include:

1. Research into the possibilities of the stimulation of visual, olfactory and taste receptors, as well as nerve fibers and central nervous system structures.
2. Use of focused ultrasound for remote stimulation of acupuncture points (corporeal and auricular) and the comparative research into these effects and those after the use of needles.
3. Comparison of brain-evoked potentials for different sensations (tactile, pain, hearing and so forth) with the aim of defining the modality of the sensation by means of the evoked potentials.
4. Comparative research of other characteristics of different skin sensations following the stimulation of the same part of the skin (thresholds, summation characteristics, locality, irradiation of the sensation and so forth).
5. Investigations of peculiarities of deep pain and other sensations in the periosteum, joints, pleura and so forth.
6. Research into the sensitization or inhibition of the action of the carrier frequency of ultrasound.
7. Investigations of sensations arising after the action of a number sequential stimuli of focused ultrasound at neural structures and research into the summation phenomena.
8. Interaction between the different receptive structures or systems during the use of several focused ultrasound transducers.
9. Synchronization of the stimuli action with definite biological cycles, rhythms and so forth.
10. Research into the physical and physiological mechanisms of focused ultrasound stimulation with the aim of improving its effective and safe application in clinical practice and in physiological research.

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## REFERENCES

Altenberg K, Kästner S. Demodulation von Ultrashallwellen in Flussigkeiten. Ann Phisik 1952;11:161–165.
Ashkinazi IA, Ishinova VA, Tsirulnikov EM. Skin sensitivity to pain under the chronic psycho-emotional stress in man. Neirophysiologija 1992;24:535–542.
Bergmann L. Der Ultrashall und seine Anvendung in Wissenschaft und Technik. Zurich: Hirzel, 1954.
Broun GR, Gavrilov LR, Zhadan GG, Ilyinskii OB, Tsirulnikov EM. Effect of focused ultrasound on the electroreceptor system of skates and upon several tissues of fishes and amphibians. J Evol Biochem Physiol (Zh Evol Biokhim Fiziol) 1980 (1981); 16:263–268.
Foster KR, Wiederhold ML. Auditory responses in cats produced by pulsed ultrasound. J Acoust Soc Am 1978;63:1199–1205.
Fry FJ. Intense focused ultrasound: Its production, effects and utilization. In: Ultrasound: Its application in medicine and biology (pt. 2). New York: Elsevier, 1978:689–736.
Fry FJ, Ades HW, Fry WJ. Production of reversible changes in the central nervous system with ultrasound. Science 1958;127:83–84.
Fry WJ. Electrical stimulation of brain localized without probes—theoretical analysis of proposed method. J Acoust Soc Am 1968;44:919–931.
Fry WJ, Mosberg WH, Barnard JW, Fry FJ. Production of focal destructive lesions in the central nervous system with ultrasonics. J Neurosurg 1954;11:471–478.
Fry WJ, Wulff VJ, Tucker D, Fry FJ. Physical factors involved in ultrasonically induced changes in living systems: 1. Identification of non-temperature effects. J Acoust Soc Am 1950;22:867–876.
Gavrilov LR. Physical mechanism of the lesion of biological tissues by focused ultrasound. Sov Phys-Acoust 1974;20:16–18.
Gavrilov LR. Use of focused ultrasound for stimulation of nerve structures. Ultrasonics 1984;22:132–138.
Gavrilov LR, Dmitriev VN, Solontsova LV. Use of focused ultrasonic receivers for remote measurements in biological tissues. J Acoust Soc Am 1988;83:1167–1179.
Gavrilov LR, Gersuni GV, Ilyinski OB, et al. The effect of focused ultrasound on the skin and deep nerve structures of man and animal. Prog Brain Res 1976a;43:272–292.
Gavrilov LR, Gersuni GV, Ilyinski OB, Tsirulnikov EM. A study of reception with the use of focused ultrasound. I. Effect on the skin and deep receptor structures in man. Brain Res 1977a;135:265–277.
Gavrilov LR, Gersuni GV, Ilyinski OB, Tsirulinkov EM, Shchekanov EE. Reception and focused ultrasound. Leningrad: Nauka, 1976b [in Russian].
Gavrilov LR, Gersuni GV, Ilyinski OB, Tsirulnikov EM, Shchekanov EE. A study of reception with the use of focused ultrasound. II. Effects on the animal receptor structures. Brain Res 1977b;135:279–285.
Gavrilov LR, Gersuni GV, Pudov VI, Rozenblyum AS, Tsirulnikov EM. Human hearing in connection with the action of ultrasound in the megahertz range on the aural labyrinth. Sov Phys-Acoust 1980;26:290–292.
Gavrilov LR, Kovalev VA, Tsirulnikov EM. Application of focused ultrasound for stimulation of the central nervous structures of invertebrates. Sov Phys-Acoust 1978;24:231–234.
Gavrilov LR, Pudov VI, Rozenblyum AS, et al. On the use of focused ultrasound to induce an auditory information to the human internal ear. Akust Zh 1977c;23:557–560 [in Russian].
Gavrilov LR, Tsirulnikov EM. Focused ultrasound in physiology and medicine. Leningrad: Nauka, 1980 [in Russian].
Godovanik OO, Gavrilov LR, Ilyinski OB, Tsirulnikov EM, Shchekanov EE. The use of focused ultrasound in the study of tactile sensitivity in neurological patients. Zh Nevropatol Psikhiatr 1978;78:1189–1192 [in Russian].
Hill CR. Optimum acoustic frequency for focused ultrasound surgery. Ultrasound Med Biol 1994;20:271–277.
Kossoff G. Analysis of focusing action of spherically curved transducers. Ultrasound Med Biol 1979;5:359–365.

Lansman JB, Hallam TJ, Rink T. Single stretch-activated ion channels in vascular endothelial cells as mechanoreceptors? Nature 1987;325:811–813.

Magee TR, Davies AH. Auditory phenomena during transcranial Doppler insonation of the basilar artery. J Ultrasound Med 1993;12:747–750.

Mihran RT, Barnes FS, Wachtel H. Transient modification of nerve excitability *in vitro* by single ultrasound pulses. Biomed Sci Instrum 1990;26:235–246.

NCRP Report No. 74. Biological effects of ultrasound: Mechanisms and clinical implications. National Council of Radiation Protection and Measurements. Washington, DC: NCRP Publications, 1983.

Nyborg WL. Optimization of exposure conditions for medical ultrasound. Ultrasound Med Biol 1985;11:245–260.

Olesen SP, Clapham DE, Davies PF. Haemodynamic shear stress activates a $K^+$ current in vascular endothelial cells. Nature 1988;331:168–170.

Pond J. The role of heat in the production of ultrasonic focal lesions. J Acoust Soc Am 1970;47:1607–1616.

Rinaldi PC, Jones JP, Reines F, Price LR. Modification by focused ultrasound pulses of electrically evoked responses from an *in vitro* hippocampal preparation. Brain Res 1991;558:36–42.

Sanghvi NT, Hawes RH. High-intensity focused ultrasound. Gastroint Endosc Clin NA 1994;4:383–395.

Terenius L. Biochemical mediators in pain. Triangle 1981;20:19–26.

Tsirulnikov EM. Physiological and clinical approaches to mechanoreception. Sensory Syst 1993;7:168–175.

Tsirulnikov EM, Gavrilov LR, Godovanik OO, et al. Determination of tactile thresholds in humans using focused ultrasound and morphological specificity of tactile reception. Fiziolog Cheloveka 1982;8:542–546 [in Russian].

Tsirulnikov EM, Gavrilov LR, Vasilyeva NN, Shatilov MS. Skin reception connected with pain. Fiziolog Cheloveka 1985;11:997–1000 [in Russian].

Tsirulnikov EM, Gurgenidze AG. Post-stimulus dermal pain. J Evol Biochem Physiol 1990;26:214–219.

Tsirulnikov EM, Gurgenidze AG, Vartanyan IA, Gavrilov LR. Special features of the tactile and pain perceptibility in the acupuncture points. Fiziol Cheloveka 1986;12:414–419 [in Russian].

Tsirulnikov EM, Kudinov VA, Monakhov KN, Raznatovsky IM. Tactile sensitivity in skin diseases of various origin. Vestn Dermatol Venerol 1988a;12:11–14 [in Russian].

Tsirulnikov EM, Vartanyan IA, Gersuni GV, et al. Use of amplitude-modulated focused ultrasound for diagnosis of hearing disorders. Ultrasound Med Biol 1988b;14:277–285.

Tsirulnikov EM, Vartanyan IA, Shcherbakova IY, Voinova LE, Sokolov KN. Tactile sensation as a part of the kinesthetic system (comparative clinical–physiological approach). J Evol Biochem Physiol 1990 (1991);26:602–606.

Vartanyan IA, Gavrilov LR, Gersuni GV, Rozenblyum AS, Tsirulnikov EM. Sensory perception. Research with the use of focused ultrasound. Leningrad: Nauka, 1985 [in Russian].

Vartanyan IA, Gavrilov LR, Svetlogorskaya ID, et al. Influence of the stimulus duration of focused ultrasound on the skin perceptibility. Sensory Syst 1990;4:52–59 [in Russian].

Vartanyan IA, Gavrilov LR, Zharskaya VD, Ratnikova GI, Tsirulnikov EM. The stimulating effect of focused ultrasound on the auditory nerve fibers of the frog *Rana temporaria*. J Evol Biochem Physiol (Zh Evol Biokhim Fiziol) 1981 (1982);17:335–341.

Wall PD, Tucker D, Fry FJ, Mosberg WH. The use of high intensity ultrasound in experimental neurology. J Acoust Soc Am 1953;25:281–285.

Wells PNT. Biomedical ultrasound. London: Academic Press, 1977.

Wright A. Investigation of a new algesimetric technique (Ph.D. thesis). Belfast, Northern Ireland: The Queen's University of Belfast, 1989.

Wright A, Davies II. The recording of brain evoked potentials resulting from intra-articular focused ultrasonic stimulation: A new experimental model for investigating joint pain. Neurosci Lett 1989;97:145–150.

Wright A, Davies II, Riddell JG. Intra-articular ultrasonic stimulation and intracutaneous electrical stimulation: Evoked potential and visual analogue scale data. Pain 1993;52:149–155.