Effect of the cavitation suppression technique during high-intensity focused ultrasound on liver tissue

（周波数変調波を用いた集束超音波による肝腫瘍治療の基礎的検討）

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Abstract

High-intensity focused ultrasound (HIFU) relies on the absorption of ultrasound irradiation to non-invasively cause thermal ablation of a tumor. We developed the cavitation suppression technique (CAST) to minimize the risks of non-target tissue damage during HIFU. This study examined the accurate focusing of the ultrasound beam to achieve precise locational targeting. In excised livers, HIFU was used to ablate specific areas of tissue. Although the theoretical volume of the ablated tissue was 1000 mm3, the volume without CAST use was 2641 ± 671 mm3; using CAST, the volume was 1148 ± 356 mm3. The deviation of the focal points in the direction of the ultrasound transducer, without CAST, was 5.0 ± 1.7 mm, but was reduced to 1.2 ± 1.5 mm when CAST was used. The technique was used to ablate hepatic tumors in rabbits, resulting in the ablated area being clearly demarcated from the adjacent, normal liver tissue. The tumor was completely ablated and necrotized. Thus, the new technique CAST may enable more accurate clinical use as a next-generation HIFU therapy for liver tumors.

1. Introduction

In principle, High-intensity focused ultrasound (HIFU) therapy depends on the ability of tissues to absorb ultrasonic energy. By sharply focusing ultrasonic energy on a target tissue such as a tumor inside the body, the tissue adjacent to the focal point is heated to≧ 80℃, causing its degeneration and necrosis within a short period time. If radiation is performed as a series of treatments, the entire cancerous area can be ablated and necrotized.

Intensive ultrasound to the body has been used for more than 80 years [1]. Fry et al. first studied the application of intensive ultrasound for the treatment of intracranial lesions in the 1940s [2]. A group at Indiana University has studied this method for more than 50 years and coined its name. Since the 1990s, HIFU has been applied to the transrectal treatment of prostatic hyperplasia and prostate cancer [3].

Subsequently, the use of ultrasound ablation has been implemented more broadly, including for the treatment of breast, renal, liver, and other cancers [4, 5].

However, High-intensity focused ultrasound (HIFU) has not been established as a general therapeutic method, as many reports of the occurrence of complications have been published [6-9].

During an ultrasound procedure, cavitation is caused in the media or tissue by the negative pressure generated by the ultrasound beam. Cavitation results in an increase in the tissue temperature in the vicinity of the therapeutic ultrasound path, as a result, local control of the ablation area is known to be reduced [10-13]. Single-frequency waves enlarge microbubbles in an area irradiated by ultrasound, and these microbubbles impede the precise focusing of the ultrasound beam on the target area. In resolution of this issue, frequency modulation waves were found to eliminate these microbubbles. Thus, a new technique was developed combining the use of single frequency (SF) waves and frequency modulation (FM) waves (Figure 1) [14]. This new technique, called the cavitation suppression technique (CAST), allows for alternating single SF waves and FM waves from the ultrasonic source. Thus, we propose CAST as a solution for the inadvertent ablation of non-target tissues and examined the effects of this technique.

2. Materials and methods

2.1 Ultrasonic exposure system

The devices used are schematically shown in Figure 2.

The applicator is composed of a spherical piezoceramic transducers (diameter, 100 mm; curvature, 120 mm; resonant frequency, f0 = 1.60-1.63 MHz), a 5-MHz diagnostic central probe (TOSHIBA, Tokyo, Japan), and a bag filled with degassed water (Figure 3). The transducer is driven by an ENI-A300 (Electronics & Innovation, NY, US) or A66-3401 (THAMWAY, Shizuoka, Japan) power amplifier through an impedance matching circuit. The maximum driving power is at least 2.5 kW, and the peak intensity at the focal point exceeds 46 kW/cm2. By rotating the central probe, a target area can be displayed on any section of the diagnostic ultrasound system. Both the positioning of the focal point, the actual scanning is performed using the XYZ automatic stage, with the overall system being controlled by a personal computer.

Conditions for CAST were :

Sweeping direction of frequency (SD) : from low to high

FM range : $\Delta f/fo = 0.3$ (30% of resonant frequency fo)

SF driving time : To = 1 ms

FM driving time : Tm = 5 ms

Length of an FM cycle : Tfm = 1 ms

Figure 1 shows a schematic principle of the CAST [14].

2.2 Temperature measurement experiment

In this experiment, we used male, Japanese white rabbits (Takasugi, Chiba, Japan) weighing 3.0-3.5 kg. During these initial experiments, the livers of these animals were exposed to HIFU, via a laparotomy. The rabbits were first anesthetized, by intramuscular-administration of ketamine hydrochloride (35 mg/kg) and xylazine hydrochloride (5 mg/kg).

The anesthetized animals were placed in the supine position, and the peritoneal cavity was opened through a subxiphoid, midline abdominal incision. After removal of the hepatic ligamentous attachments, the subphrenic space was filled with gauze sponges to achieve better liver exposure.

The normal rabbit livers were exposed to HIFU with or without CAST at a power of 400 W and a focal intensity of 8,000 W/cm2 for 1 s, and the temperature at the target site and surrounding tissue was measured with a platinum resistance thermometer (0.2 mm in diameter). The target point was identified as peak by weak irradiation. The temperature measuring at each point was performed five times and the data have been plotted at the mean values of the temperature.

2.3 Locations and volumes of ablated areas of excised livers

We irradiated large areas of porcine livers and then examined the sizes of the ablated areas and the gaps between the focal points and the ablated areas.

For this experiment, fresh porcine livers were purchased from a butcher shop.

In order to achieve extensive irradiation, each HIFU irradiation area was slightly shifted to obtain a steric ablated area composed of several overlapping irradiation areas. The high-temperature area was cigar-shaped, 10 mm in height and 2 mm in width, by one exposure [14]. We performed 25-point irradiation, arranged in a 5 × 5 exposure pattern. The exposure separation was set at 2 mm. The shape that was planned to ablate was a 10 mm cube (Figure 3).

In this experiment, slight shifts of irradiation areas were obtained by moving the transducer along the x- and y-axes employing the three-dimensional gantry system.

The focal plane was set at 15 mm below the surface of the liver, to ensure that the expected ablated area would fit entirely within a sample.

HIFU (power, 400 W; focal intensity, 8,000 W/cm2) was applied to for 1 s with or without CAST. The interval between shifting the transducer to the next location and the start of the next irradiation was set at 3 s.

After HIFU irradiation, the livers were examined macroscopically. In order to calculate the volume of ablated tissue, the livers were sliced into 2-mm sections along the irradiation axis, and the length of the ablated areas was measured. The middle of an ablated region in the depth direction was regarded as a focal point of irradiation.

2.4 Irradiation to of solitary VX2 tumors

Japanese white rabbits with VX2 tumors were purchased from Funabashi Farm (Chiba, Japan), and the VX2 carcinomas were implanted into the femoral muscles of other Japanese white rabbits (Takasugi). Solitary hepatic VX2 tumors were created by implanting the tumor pieces in the medial lobe of the liver.

The hepatic tumors were displayed on the ultrasonic diagnostic device for size measurements. An irradiation area, including the tumor and an adequate margin of surrounding tissue, was determined by computer coordination. Tumors of nearly all sizes and forms were found to be capable of being irradiated by this method.

For this experiment, animals were prepared and anesthetized employing the same procedure as for **Temperature measurement experiment**. Multiple-point HIFU irradiation was applied to the livers at a power of 400 W for 1 s at 3-s intervals with the CAST method. The rabbits were sacrificed 24 h after irradiation, and their livers were excised for examination. After formalin fixation, the liver specimens were sliced into sagittal sections, and microscopic sections were stained with hematoxylin-eosin.

3. Results

3.1 Temperature measurement experiment

The horizontal and vertical distributions of temperatures around the focal points are shown in **Figure 4** and **5**, respectively.

On the horizontal plane, the highest temperature was at least 80℃, regardless of the use of CAST. The temperature was the same at the center point and was high enough to ablate the target tissue. According to the vertical distribution of temperatures, the point with the highest temperature, in the absence of CAST, was located 5 mm in front of the predicted site (focal point). However, this deviation was reduced to 2 mm with the use of CAST. Although the area without CAST use deviated markedly in the direction of the transducer, the high temperature area with CAST use was essentially consistent with the target area.

3.2 Locations and volumes of ablated areas in excised livers

The ablated areas were apparent by macroscopic examination and were hard when palpated. The areas were white and had clearly defined borders distinguishing them from the untreated, normal parts of the liver.

The volumes of the ablated areas, when CAST was employed, are shown in Figure 6. The volume without CAST use was 2641 ± 671 mm3; with the use of CAST, the volume was 1148 ± 356 mm3, which was approximately equal to the theoretical value.

The deviations of the focal points in the direction of the transducer, when CAST was used, are shown in Figure 7. Although the deviation, without CAST use, was 5.0 ± 1.7 mm, when CAST was used, the deviation was reduced to 1.2 ± 1.5 mm.

An actual photographic images of these ablated areas are shown in Figure 8. Although the ablated area, without the use of CAST, was expanded and deviated in the direction of the liver surface, the area and size of ablation with CAST use were achieved essentially as planned.

3.3 Irradiation of solitary VX2 tumors

Tumors were visualized by diagnostic ultrasound, and their sizes were measured. Because the size of the ablated area depends on the number and pattern of exposures, the irradiated area included the tumor and an adequate margin of surrounding tissue. HIFU irradiation was applied with a 7 × 10 exposure pattern.

The ablated area was macroscopically apparent and included the tumor. In the histological study, the ablated area was clearly demarcated from the adjacent, unablated normal liver tissue. The tumor was completely ablated and necrotized (Figure 9).

4. Discussion

HIFU has the ability to selectively ablate tissue targets without damaging the surrounding tissues, i.e., tissues in front of and behind the target, by focusing the ultrasound beam on an extremely small target area.

In recent years, the development of minimally invasive therapy (MIT) has been increasingly sought in response to patients' growing demands for quality of life (QOL), therefore HIFU is expected to be an ideal local therapy.

Based on the shape of the applicator, HIFU can be classified into two types, the transrectal type and the transabdominal type. Both types are clinically used in a limited number of institutions; the transrectal type is used for the treatment of prostate diseases and the transabdominal type for the treatment of uterine myomas. The technique involves partial irradiation of a large target, and is mostly used as a lesion reduction therapy. HIFU treatment is only covered by health insurance for benign prostatic hyperplasia in authorized medical facilities in Japan, and it is not in wide use elsewhere in the world. The indication of HIFU for diseases in other regions such as liver is still performed in clinical trials [15, 16].

Hepatic tumors have been treated in various ways such as surgical resection, transcatheter Arterial Chemoembolization (TACE), percutaneous ethanol injection therapy (PEIT), and thermal ablation using microwaves and radiofrequency emitted from a needle electrode. We have been engaged in the development of radiofrequency ablation as a type of thermal ablation for liver tumors [17-19]. During the process, we realized that clinically, thermal ablation had anti-neoplastic effects on liver tumors. In 1995, considering that HIFU, which does not require insertion of electrodes, is also a minimally invasive, local, anti- neoplastic treatment, we began conducting studies on the efficacy of HIFU for treatment of liver tumors [20, 21]. Because a high level of energy is transmitted into the body within a short period of time, destruction of tissue in an unplanned area is possible and may cause serious complications. When HIFU irradiation is performed expecting complete cure of malignant tumors, such as liver cancer, an area wider than the extent of tumor needs to be irradiated, which raises the concern of a higher risk. Therefore, in order for HIFU to be used for the treatment of malignant tumors, more focused and accurate irradiation is needed.

However, it is difficult to focus the ultrasound beam accurately on the target area in HIFU therapy. Heating of the target by the ultrasound waves is reduced by the presence of cavitation in the irradiation area; this would result in the heating region extending and moving to the transducer side; we encountered such result, and similar results have also been reported [22, 23]. This difficulty of focus control with cavitation leads to the occurrence of complications and is still a problem [24-27]. HIFU is known to induce thermal damage in off-target tissue, with this being especially problematic for superficially situated tumors, in which burn complications on overlying structures are easily induced [28]. The problem of cavitation has not been resolved, however, by taking advantage of ablation of a wider area than expected, attempts are underway to reduce treatment time [29, 30].

Our study is the first attempt to improve the local control capability by suppressing cavitation. A new technique called CAST utilizing FM driving was proposed, which enables the ultrasound energy to reach the focal region and reduces the near-field heating in front of the focal spot during focus scanning by suppressing cavitation along the ultrasonic path. The present study showed that the use of

CAST resolves the problems associated with the deviation of focal points and ablated areas to improve local control. This study demonstrated that a liver tumor, observed with an ultrasonic diagnostic device, may be simultaneously and precisely treated in a minimally invasive manner. Thus CAST is thought to be useful for protecting the surrounding tissues from any adverse effects that could be caused by shifts in the thermally coagulated region.

However, there are specific issues that need to be resolve when this device is used for extracorporeal irradiation of liver tumors. HIFU is performed through an intercostal space or from below the costal arch, depending on the location of the tumor.

Because bones strongly reflect ultrasound, the presence of a rib cage is a significant hindrance to the potential applications of focused ultrasound as a noninvasive extracorporeal surgery modality [31]. When the rib cage is in the HIFU propagation path, it will also be necessary to calculate the shielding ratio of ultrasound and compensate for this shortfall by increasing the intensity of irradiation energy [32]. HIFU irradiation using a transabdominal applicator is easily applied to uterine myomas as it can be used from the lower abdomen, where no consideration of costal bone shielding is required. In addition, as it is a benign disease, even if some tissues are not cauterized, symptom alleviation alone may be acceptable.

Furthermore, due to the physical characteristics of ultrasound, the absorption coefficients of irradiated tissue vary greatly among tissue types. Even absorption coefficients for the same type of tissue vary among individuals. Because the tissue thicknesses and shielding ratios vary between individuals, an accurate calculation of the energy necessary for irradiating the tumors by either method prior to the actual irradiation procedure is difficult.

Despite these issues, HIFU is currently being studied at many institutions, with the anticipation that these challenges will be resolved.

HIFU, as a new thermal therapy, non-invasively induces complete coagulative necrosis of a tumor without surgical exposure or insertion of instruments into the lesion. The burden on patient is small, and there is no issue of resistance. Moreover, this technique is expected to provide favorable functional preservation of the organ without resection and avoid intraoperative proliferation or metastasis of tumor cells. These advantages make it radically different from the conventional therapies and one of the most attractive options for the local therapy of liver tumors.

We believe that with advances in research and development on irradiation techniques, HIFU has the potential to spread rapidly.

5. Conclusions

HIFU requires no incision or puncture, can be performed repeatedly and appears to be highly attractive as a minimally invasive treatment for liver tumors.

In this study, our newly developed device CAST - a new technique alternately driven by single frequency waves and frequency modulated waves - improved local control and allowed the accurate, non-invasive ablation of the target areas of rabbit and porcine livers. Our results lead to the possibility that in clinical practice, a liver tumor detected by an ultrasonic diagnostic equipment may be treated safely with HIFU in a minimally invasive manner.

Thus, the new technique CAST may enable more accurate clinical use as a next-generation HIFU therapy for liver tumors.

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Figure Legends

Figure 1: Cavitation Suppression Technique (CAST). CAST is a technique combining single - frequency waves and frequency modulation waves.

Figure 2: Schematic diagram of the experimental system.

Figure 3: Twenty-five point irradiation pattern arranged in a 5×5 alignment of exposures.

Figure 4: Horizontal distributions of temperatures around the focal point with and without the cavitation suppression technique (CAST).

Figure 5: Vertical distribution of temperatures around the focal point with and without the cavitation suppression technique (CAST).

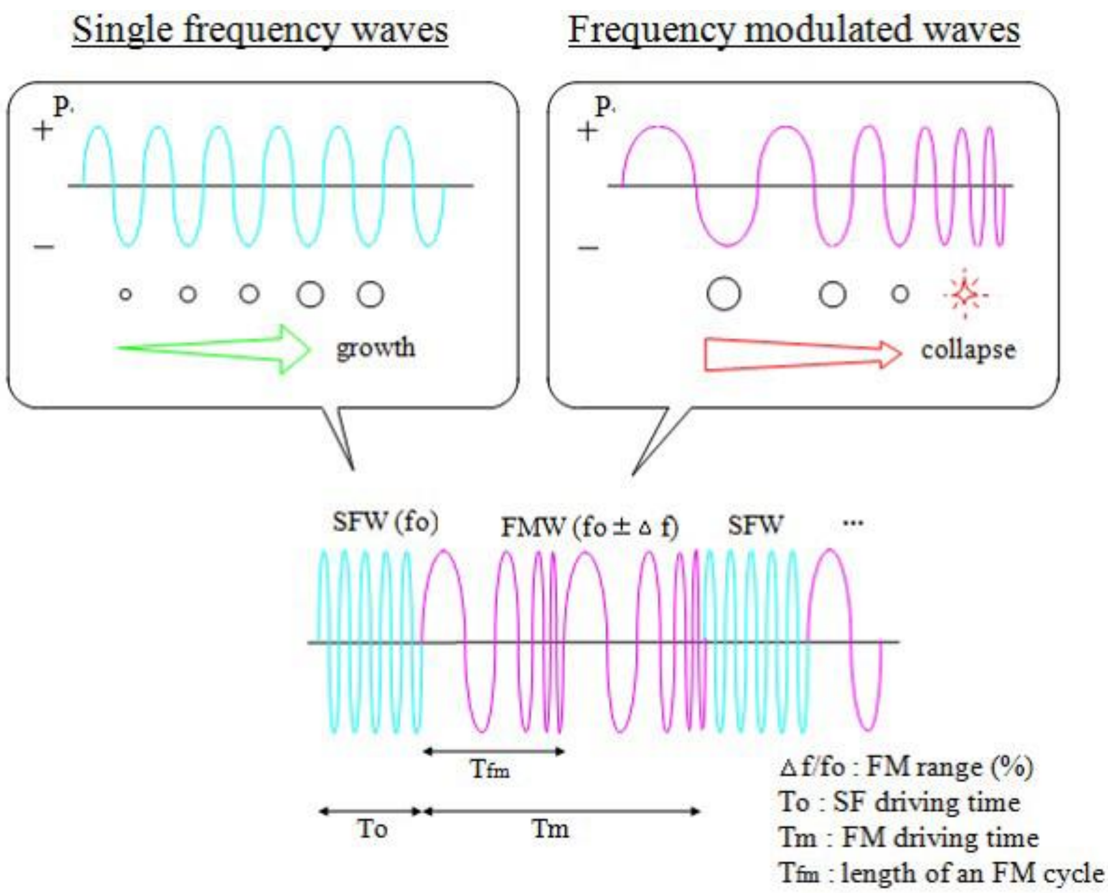
Figure 6: Volumes of the ablated areas with and without the cavitation suppression technique (CAST).

Figure 7: Deviations of the focal points with and without the cavitation suppression technique (CAST).

Figure 8: Volumes of the ablated areas with and without the cavitation suppression technique (CAST).

Figure 9 Photographic images after high - intensity focused ultrasound irradiation of solitary VX2 tumors.

Figure 1


Single frequency waves
Frequency modulated waves
P
+
−
growth
collapse
SFW (fo)
FMW (fo ± Δ f)
SFW
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Tfm
To
Tm
Δ f/fo : FM range (%)
To : SF driving time
Tm : FM driving time
Tfm : length of an FM cycle

Figure 2

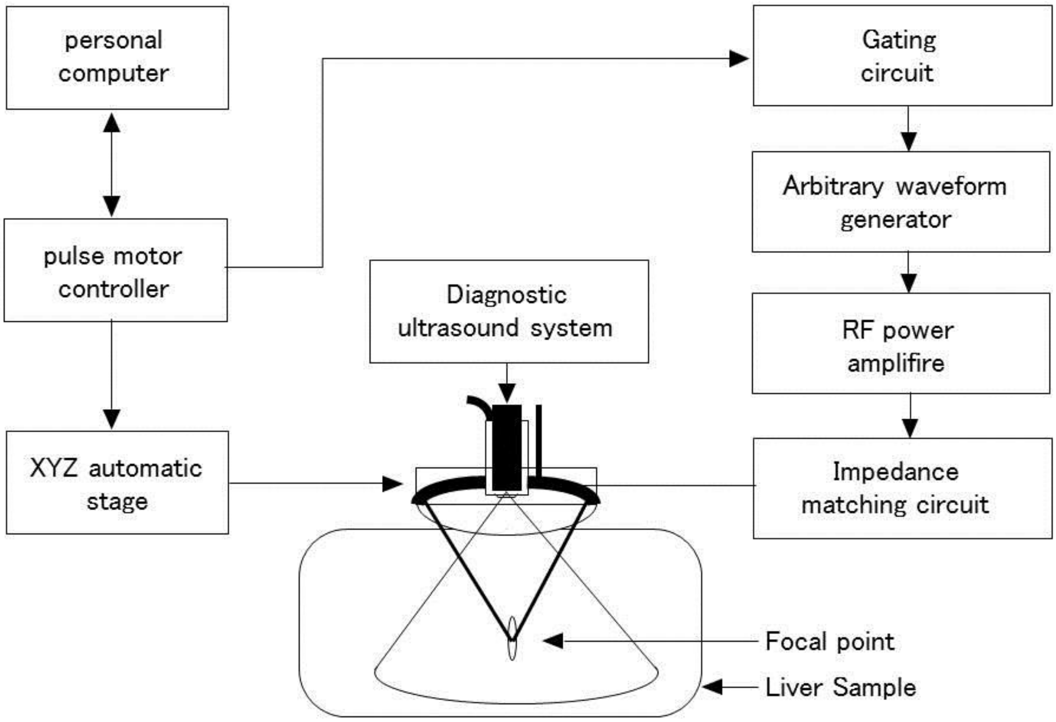

personal computer
pulse motor controller
XYZ automatic stage
Gating circuit
Arbitrary waveform generator
RF power amplifire
Impedance matching circuit
Diagnostic ultrasound system
Focal point
Liver Sample

Figure 3

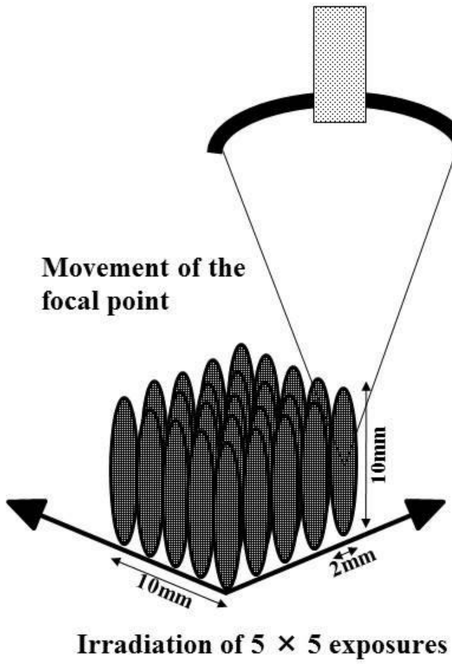

Movement of the
focal point
10mm
10mm
2mm
Irradiation of 5 × 5 exposures

Figure 4

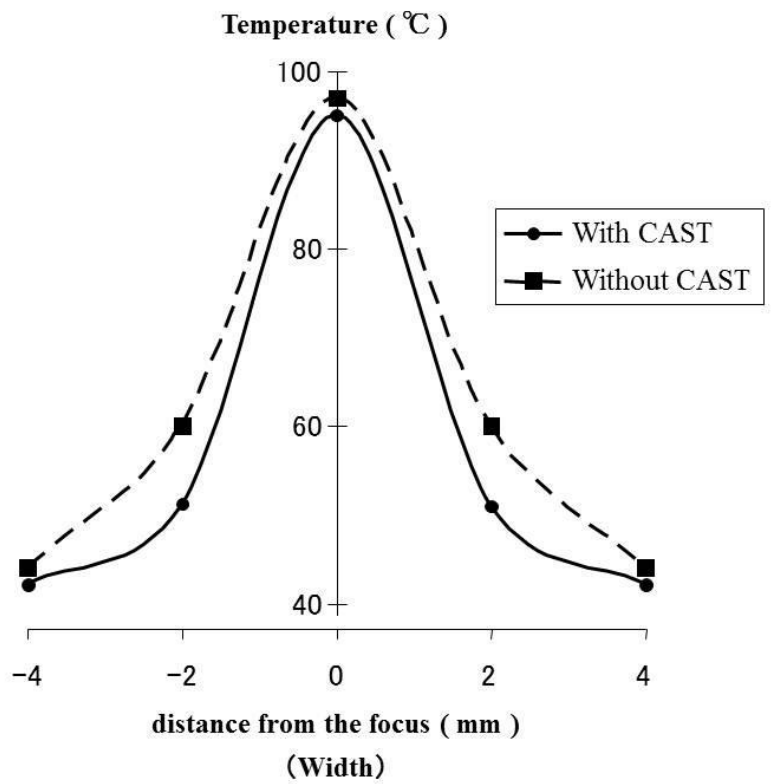
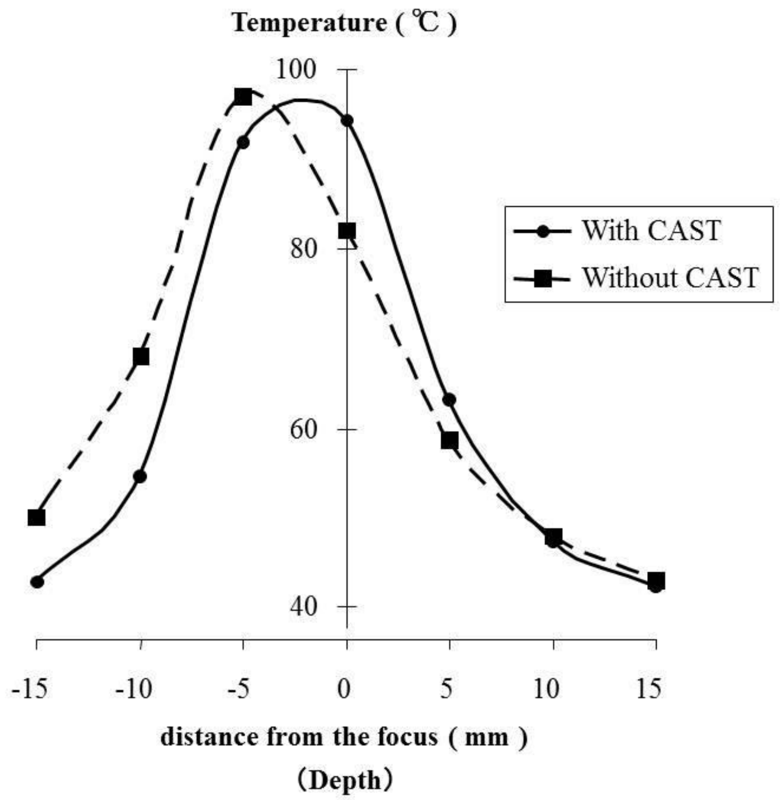

Temperature (℃)
100
80
60
40
With CAST
Without CAST
−4
−2
0
2
4
distance from the focus (mm)
（Width）

Figure 5



Temperature (℃)
100
80
60
40
With CAST
Without CAST
-15
-10
-5
0
5
10
15
distance from the focus (mm)
(Depth)

Figure 6

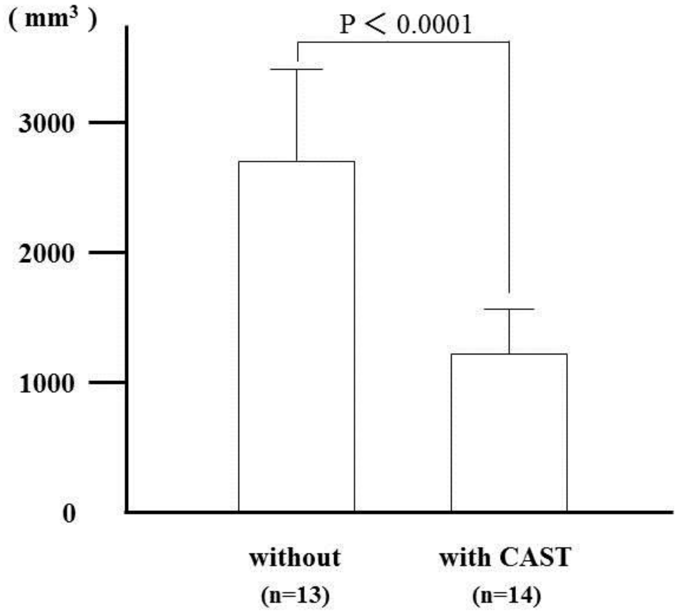


Figure 7

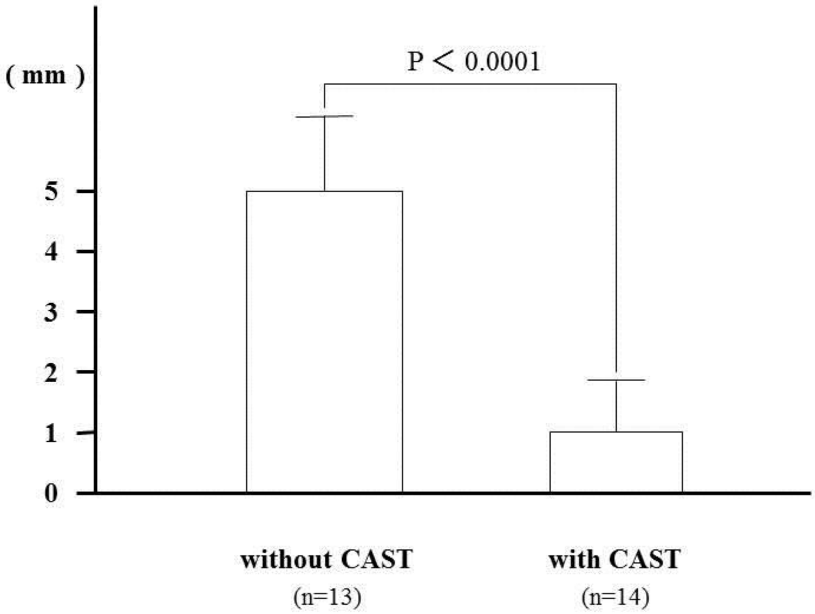
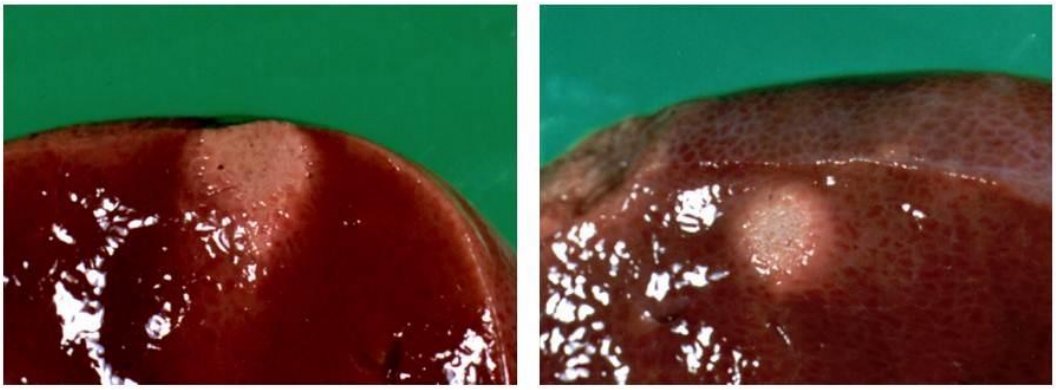
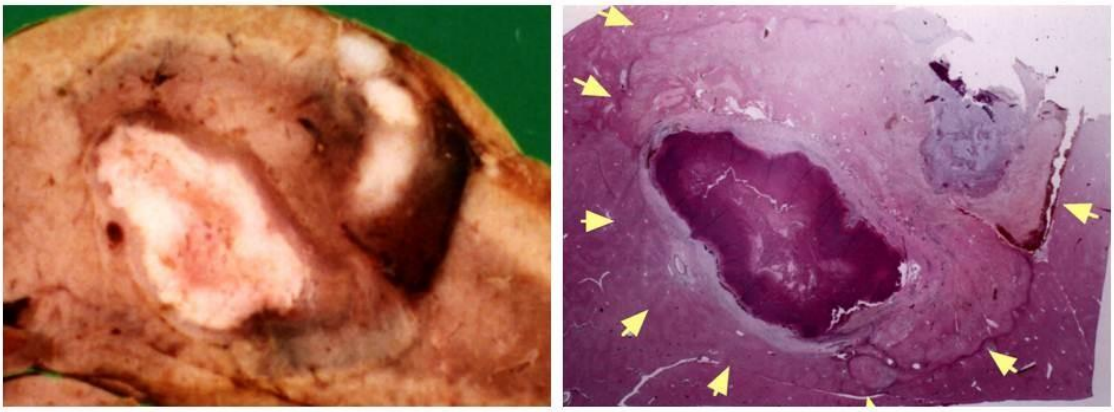


Figure 8



Without CAST **with CAST**

Figure 9



Macroscopic views **HE staining**

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