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Development of a Minimally Invasive and Non-invasive Lipolysis Laser System for Effective Fat Reduction

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Abstract

Introduction: Obesity is a global problem because it causes various complications. Methods for reducing fat for healthy life are being studied. In this study, we developed a minimally invasive and non-invasive lipolysis laser system for effective fat reduction.

Methods: The laser had the wavelengths of 1980 nm and 2300 nm which have very good absorption of fat and water. We developed a minimally invasive laser system that breaks down fat by direct irradiation of fat tissue. This minimally invasive laser system uses a 808 nm diode laser and Nd:YVO_4 to generate the 1064 nm wavelength, which is the pumping source of the nonlinear crystals. It is a mid-infrared lipolysis laser system having two wavelengths of 1980 nm and 2300 nm by controlling the temperature of nonlinear crystals. We also developed a non-invasive laser system that reduces fat with hyperthermia treatment by raising the temperature of adipocytes with a 1060 nm penetrating depth into the skin. In this non-invasive laser system, the In gallium arsenide (GaAs) diode laser is irradiated on the skin with an area of 4 × 8 cm^2 through the hand-piece. The cooling system in the hand-piece protects the skin from burns. We studied the effectiveness and safety of each system through animal experiment. We studied the effects of lipolysis when these two systems were combined.

Results: This research uses new wavelengths (1980 nm, 2300 nm) to increase the fat reduction effect with low energy (1.3 W). After using the 1060 nm (1.1 W/cm^2) wavelength laser, when the 1980 nm and 2300 nm (1.3 W) laser were used, a lipolysis effect of about 35 % was obtained.

Conclusion: We have developed a 1.3 W mid-infrared (1980 nm, 2300 nm) laser with good lipolysis effect with low power.

Keywords: Mid-infrared, Laser lipolysis, Low energy laser, Nonlinear crystal

Introduction

In modern society, obesity does not mean being overweight, but it means a condition in which too much fat is accumulated in the body. Due to changes in eating habits and lifestyles, the obese population increasing by 20%.[1] Methods for reducing fat for healthy life are being studied. In order to solve many complications caused by obesity, techniques for reducing and removing fat have been developed continuously. The method of lipolysis involves inserting a device into the body to suck up fat through a suction machine, destroying fat cells in the body using ultrasound equipment or a laser in vitro, and irradiating the laser directly to the adipose tissue through minimal invasion. There are non-invasive methods such as high frequency lipolysis, cool lipolysis, injection and drug ingestion. Laser lipolysis was first studied by Apfelberg in 1992.[2] In 2002, Neira's presentation on the effects of laser-assisted liposuction[3] led to increased interests in a laser-assisted lipolysis. After approval by the FDA in October 2006, the laser lipolysis is a commonly used method for removing superfluous fatty tissue and is being developed steadily. Laser lipolysis not only reduces fat and shortens recovery time, but also prevents bleeding by coagulating blood vessels, and it is effective for strengthening skin by inducing collagen production.[4-7] Laser lipolysis generates a single wavelength using a diode laser or a Nd:YAG laser. When the laser beam meets the tissue, the beam is transmitted, scattered, reflected and absorbed. For biological effects, the laser beam must be absorbed into the tissue. The absorption rate of the laser beam for the tissue is determined by the natural frequency of molecules constituting the tissue and the frequency of the entering laser. In order to maximize the effectiveness of laser treatment, it is important to choose a wavelength that is appropriate for the treatment purpose. The wavelengths used for lipolysis are 924, 968 and 980 nm for the diode laser and 1064, 1319, 1320 and 1440 nm for the Nd:YAG laser.[8] Fat reduction by the laser depends on

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the wavelength and energy of the laser.[9] According to the theory of selective photothermolysis, tissue preferentially absorbs the energy of the laser based on the absorption coefficient of the fat and water of the wavelength.[8] Figure 1 shows the absorption rate of water and fat with respect to the wavelength.

Recently, we have been interested in the method of lipolysis by laser irradiation on skin without invasion. This is a way of reducing fat in the body by stimulating the adipose cell layer with the principle of hyperthermia treatment that the laser energy increases the temperature of the adipocyte to 42~47°C.[10,11] As shown in Figure 2, the 1064 nm wavelength region of the Nd:YAG laser has deep penetration depth compared to other wavelengths.

Non-invasive lipolysis laser delivers the laser energy to the fat cells even when irradiated on the skin.[12] The mechanism of non-invasive laser lipolysis relies on temperature. 40~47°C is reported as the death threshold of adipocytes, and the total decomposition of adipocytes happens at 50~65 °C.[13-16] To protect the skin during laser irradiation, the hand-piece includes a cooling system. This cooling system does not have a therapeutic effect, unlike cryotherapy. The cooling system of the hand-piece allows the temperature of the skin to be 15 °C during treatment.[11] The disrupted adipocytes by the hyperthermia treatment are removed through the body's natural mechanism, and inflammation induces macrophages to remove cellular debris.[12] In this paper, we performed three studies. First, we developed a minimally invasive laser system that reduces fat by direct irradiation on fat tissue. It had wavelengths of 1980 nm and 2300 nm which are good at the absorption of fat and water. Second, we developed a non-invasive laser system that reduces fat with hyperthermia treatment by raising the temperature of adipocytes with a 1060 nm penetration depth into the skin. Third, we confirmed the efficacy and safety of each system through animal experiments and confirmed the lipolysis effects when the two systems were combined.

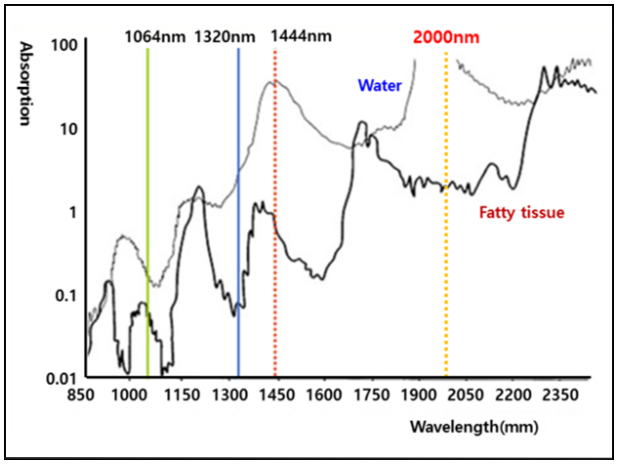

Figure 1. Comparison of the Laser Absorption Rate for Fat and Water.

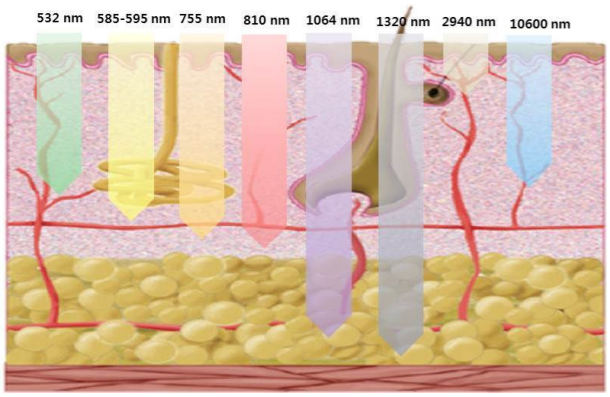

Figure 2. Depth of Skin Penetration by Wavelength.

Materials and Methods

Laser System Development

Minimally Invasive Laser System

Generally, DPSS[17] laser with the wavelength of 1064 nm uses Nd:YAG and Nd:YVO_4 as a gain medium. To make a 1064 nm infrared laser source, either flash lamp excitation method or LD excitation method is used. For the excitation method of the flash lamp, dozens of watts of high power pulse lasers have been developed. However, Faraday rotators and polarizers are used to control the polarization, which limits the size of the laser system. We used a diode pumping method for a compact laser system. In this case, higher output is obtained when Nd:YVO_4 is used as the gain medium rather than Nd:YAG.[18,19] Nd:YVO_4 has an absorption rate of 808 nm that is five times higher than Nd:YAG, which is able to extends double the lifetime of the diode laser. As the single axis crystal with large double refraction, it is possible to obtain a linearly polarized beam without a polarization device by using Nd:YVO_4 having the property of emitting a linearly polarized beam.[20] The crystal of Nd:YVO_4 is rectangular, with "a" and "c" directions orthogonal to each other. The laser rod usually orients the rod axis along an A-axis of the crystal. The Nd:YVO_4 mount with cooling system can reduce the thermal lens effect generated when focusing the 808 nm excitation beam on the Nd:YVO_4. As a result, a stable 1064 nm beam can be obtained. Sine pulsed lasers have the advantage of obtaining high peak power from low energy, AO Q-switch is used to create pulsed beams with repetition rates of tens of kHz.[21] In order to construct an intra cavity, an optical parametric oscillator composed of a mirror and a non-linear optical medium periodically polarized was placed inside the Nd:YVO_4 laser resonator. In non-linear optical crystal used as a laser, rods are changed, the length of the crystal axis changes with temperature and the output wavelength changes due to the change in the length.

As shown in Figure 3, an oven with a heater and real-

time temperature sensor was designed and made for temperature control of non-linear optical crystal for stable wavelength output. To select non-linear optical crystals to obtain wavelengths of 1980 nm and 2300 nm suitable for lipolysis, Bruner's proposed "temperature dependent Sellmeier equation" for the refractive index of SLT was used.[22,23] We calculated the temperature-dependent wavelengths of non-linear optical crystals such as CSP, PPLN, APMgLN, and PPSLT. As shown in Figure 4, we found that wavelengths of 1980 nm and 2300 nm were generated at 110°C of PPSLT.

Three wavelengths, namely 532 nm wavelength, 2300 nm wavelength and 1980 nm wavelength, can be obtained at 110°C of PPSLT. Among the wavelengths, mid-infrared wavelengths (1980 nm and 2300 nm), which are effective for lipolysis, are delivered to the fiber through dichroic filters and focusing lenses. The dichroic filter consists of three filters and a rotating motor to choose between 1980 nm and 2300 nm wavelengths respectively, or two combined wavelengths. The focusing lenses were designed and made to minimize the coupling loss when focused on fiber with 0.22 NA and 400 μm core size. Figure 5 shows the measurement results of the wavelength and power of the mid-infrared laser system for lipolysis. The temperature of non-linear optic crystal obtained wavelengths of 1980 nm and 2300 nm at 175°C. The 1980 nm and 2300 nm wavelengths are good absorption of water and fat, and the maximum power delivered to the fiber is 1.67 W. We have developed a minimally invasive mid-infrared lipolysis laser system that includes a laser head, temperature controller, power supply and cooling-system.

Non-invasive Laser System

In the non-invasive laser, externally radiated laser energy is delivered to and absorbed by the adipocytes.[16] A source used a 1060 nm wavelength semiconductor diode laser with the deepest penetration into the skin. The hand-

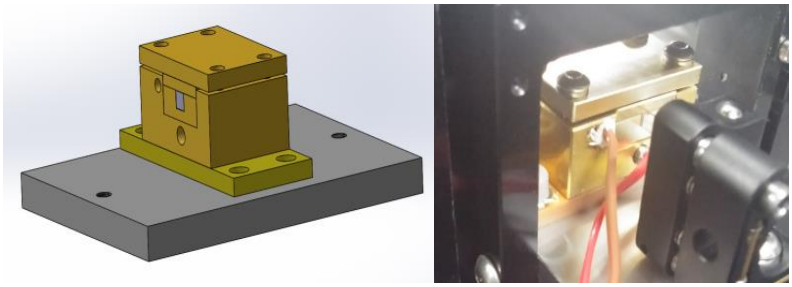

Figure 3. The Oven for Temperature Control of Non-Linear Optical Crystal, (a) Design, (b) Production and Application.

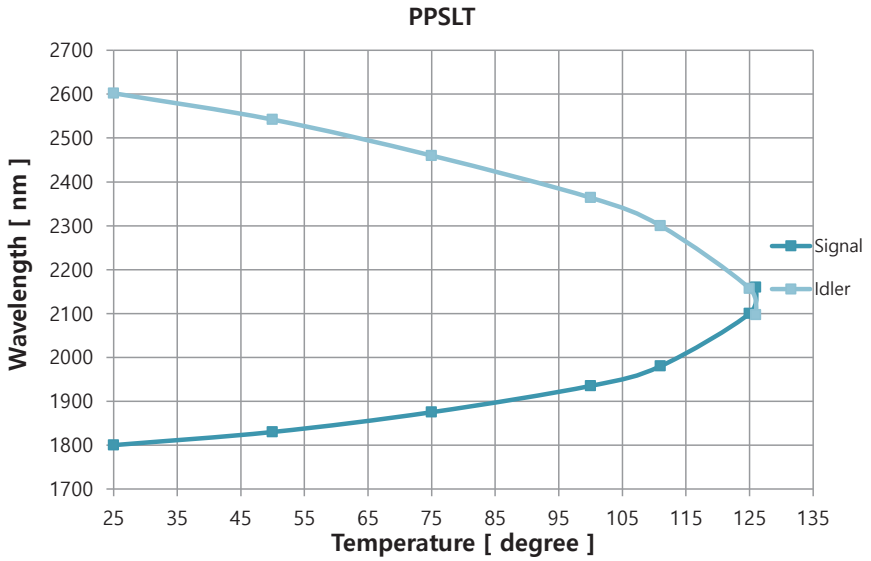

Figure 4. Variation of Wavelength of Non-Linear Optical Crystal With Temperature.

piece consists of a 1060 nm LD, optical system for the top-flat beam, cooling system and sapphire window. The rod shape LD has 40° and 80° divergence angle on the horizontal and vertical axis, respectively. The Gaussian-type beam has higher center beam intensity, and the beam intensity decreases toward the edges. The optical system is designed to increase lipolysis efficiency by irradiating a wider area with even beam intensity. In order to make the beam uniform, a cylindrical lens and a hand-piece coated with metal for diffuse reflection were used.

Results

As shown in Figure 6, the beam uniformity of the Laser Diode LD (Laser Diode) was increased to more than 85% using the designed cylindrical lens and reflector. We could maintain the temperature of skin at 16°C during irradiating the laser by using a selective cooling method. This is important because it is closely related to safety. To cool the surface of the hand-piece in contact with the skin, as shown in Figure 7, TEC (thermoelectric couple) was attached to both sides of the hand-piece and the water path was divided into two directions to increase the cooling efficiency of LD and TEC.

TEC is a cooling device using the Peltier effect, that is, the difference in voltage is converted into a difference in heat in a semiconductor bonded to metal. Figure 8 shows the results of measuring chiller temperature and TEC temperature of each hand-piece for water cooling during laser irradiation.

To keep the average temperature of the hand-piece constant, the laser is not continuously irradiated. The laser was irradiated for total of 25 minutes. The laser is not irradiated continuously for 25 minutes, the laser is irradiated for 20 seconds and 10 seconds break is repeated. As a result of the measurement during the laser irradiation, the cooling water (chiller) temperature

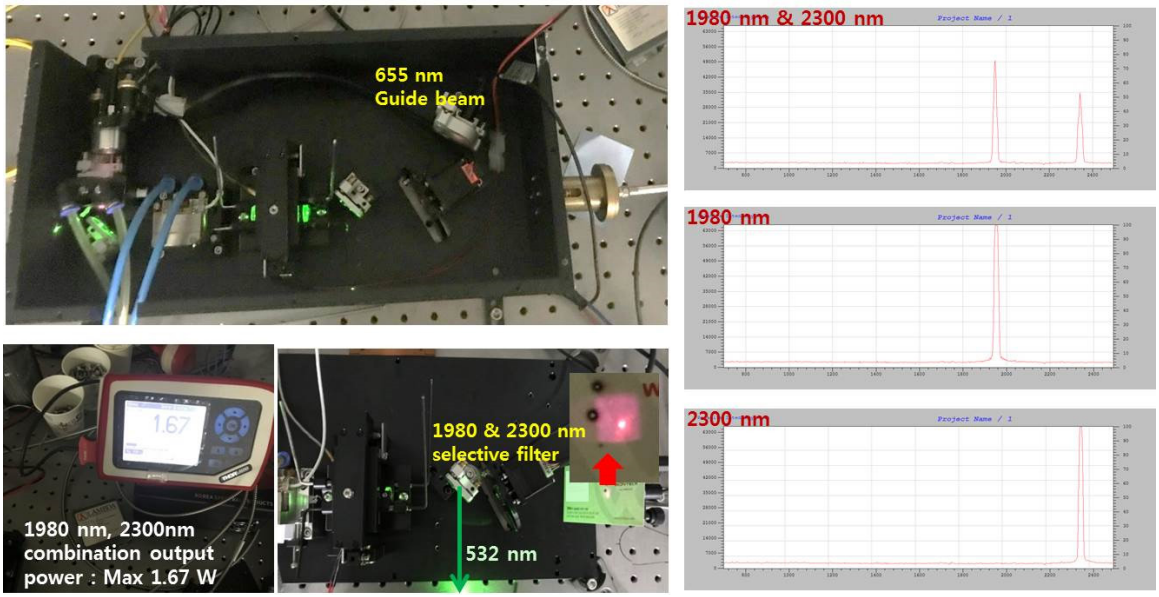

Figure 5. Output Wavelength and Power Measurement Results of the Lipolysis Laser System.

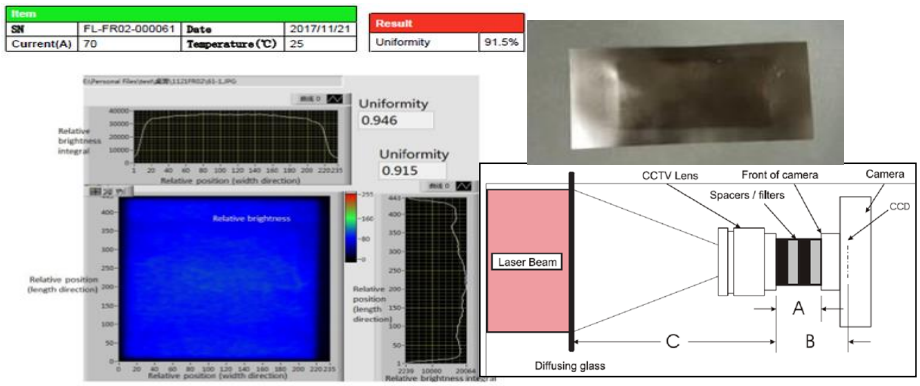

Figure 6. Results of Beam Uniformity Design and Test of the Hand-Piece.

was maintained at 21°C and the average of each TEC temperature was also maintained at 16°C. In addition, it is designed to raise the laser irradiation area to increase the laser irradiation efficiency. The irradiation area of the laser beam is developed 4 × 8 cm^2, and the irradiation area is increased by more than 30 % compare to the conventional equipment of 4 × 6 cm^2. These hand-pieces are designed to adjust each output individually or simultaneously. The maximum power of the laser is 1.4 W/cm^2 per hand-piece.

Figure 9 shows the result of measuring the output wavelength of each hand-piece. The output wavelength peak of the hand-piece was 1059 nm ± 1 nm, and the wavelength width (FWHM) was 2.3 nm at the maximum, thereby obtaining a stable wavelength. We have developed a non-invasive lipolysis laser irradiator that includes a hand-piece, LD controller, cooling system and power supply.

Lipolysis Preclinical Experiment Results

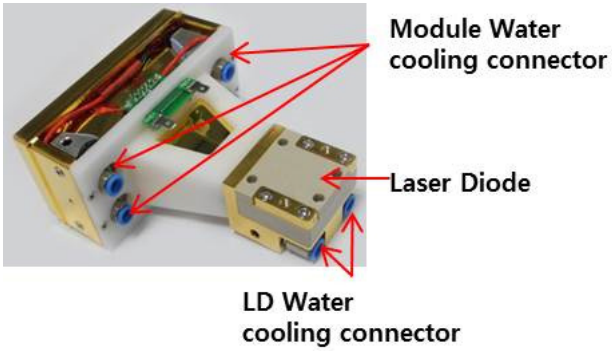

Figure 7. Hand-piece With a Cooling System

Ex-vivo and in-vivo animal experiments were performed to confirm the effective and safety of the minimally invasive mid-infrared laser system and the non-invasive laser system. Based on the results obtained in this experiment, the experiments of the two systems were combined. Animals used in the experiment were guinea pigs. Guinea pig's skin is mainly used because skin color, hair follicles, sweat glands, and subcutaneous fat are similar to humans. It is suitable for evaluating the efficacy and safety of laser therapy devices.

Minimally Invasive Laser System

Prior to the experiment, we determined the output of each wavelength by referring to a research paper authored by Kim and Kim,[17] who conducted ex-vivo experiments using the wavelengths of 1980 nm and 2300 nm. We conducted ex-vivo experiment using the guinea pig's bulk tissues. The guinea pig bulk tissues were tested after processing temperature adjustment in a 37°C incubator for 12 hours. The optical fiber is inserted into the tissue through the cannula shown in Figure 10 and the laser is irradiated at the optical fiber tip.

In order to exclude tissue damage by the cannula and check the fat cell degradation by laser irradiation, the cannula moves in a fan shape. The tissue change in the red line area at the end of laser irradiation is observed as shown in Figure 11.

The guide beam was used to visually check the position where the laser irradiated. The laser power used was irradiated with 1980 nm-0.8 W, 2300 nm-0.4 W, 1980 nm and 2300 nm combination-1.2 W for 1 minute, 2 minutes and 4 minutes, respectively. After the laser irradiation, the tissues were analyzed by H&E staining. As shown in Figure 12, when the 1.2 W of the 1980 nm and 2300 nm

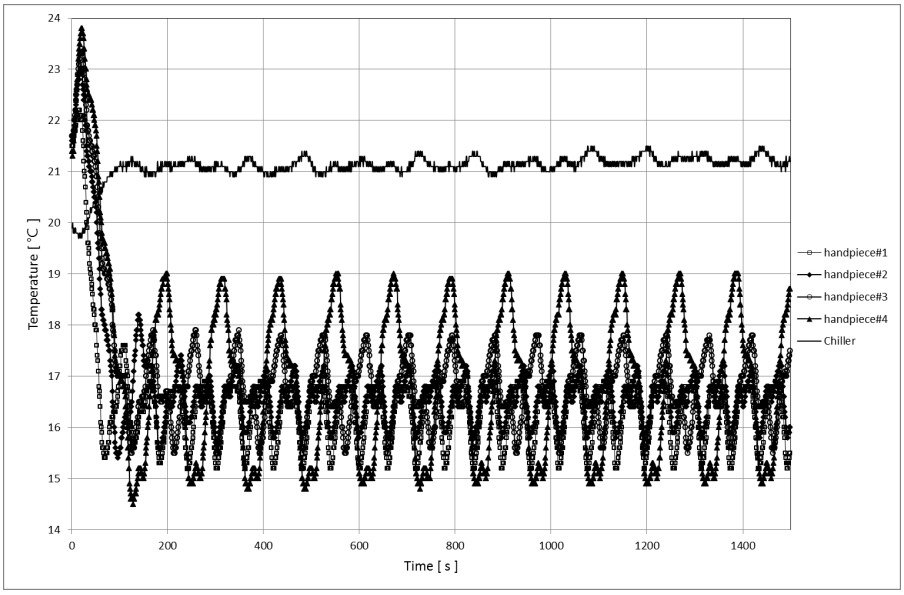

Figure 8. Cooling Temperature Measurement Results During Laser Irradiation.

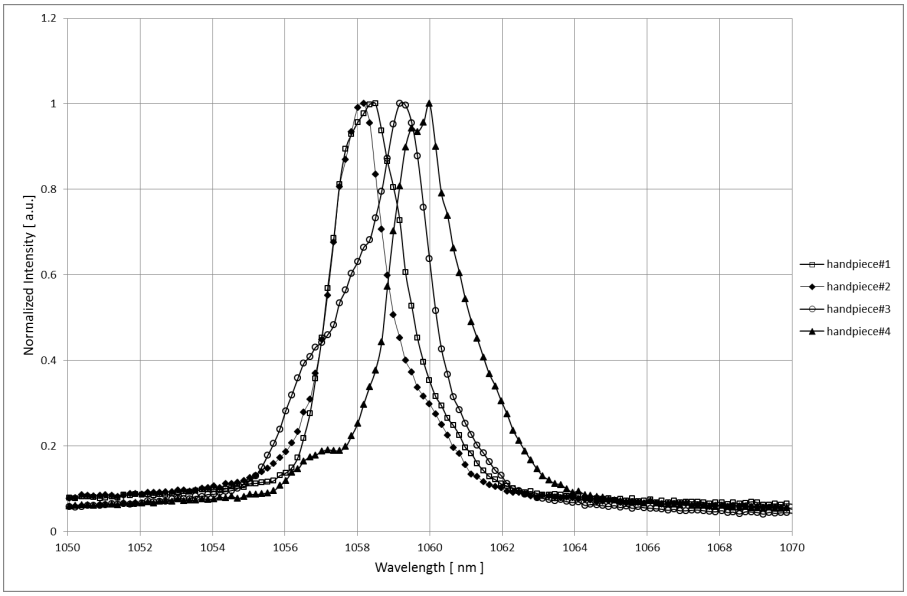

Figure 9. Output Wavelength Measurement Results of Each Hand-Piece.

Figure 10. Cannula Used to Insert Optical Fiber Into Tissue.

combination wavelengths were irradiated for 4 minutes, fat cells were decomposed (blue arrows).

We performed the in-vivo animal experiment that based on ex-vivo experimental results (Figure 12). In this experiment, one male mini-pig, 12 months old, was used. The experimental group was irradiated with the laser by dividing the pig abdomen into six parts for obtaining multiple results. In the condition of the laser irradiation, the parts of #1~#5 were irradiated with 1.2 W for 5 minutes at the combination of 1980 nm and 2300 nm wavelengths. The part of #6 was irradiated with 1.2 W for 5 minutes at 2470nm for comparison. Figure 13 shows the process of laser irradiation on the fat layer of pig abdomen. The safety was checked by visual observation of skin, blood and biopsies. The efficacy was checked by the change in the thickness of the fat layer using ultrasound. The biopsy was performed 15, 30, 60 and 90 days after laser irradiation, respectively. Blood tests were performed eight times (0, 2, 4, 8, 12, 18, 24, 48, 72 hours) for 3 days after laser irradiation. Abdominal ultrasound was performed before and after 0, 1, 7, 15, 30, 60 and 90 days.

The visual observation result could not confirm any changes and abnormalities in the laser irradiated area as shown in Figure 14.

Figure 15 is a graph showing the change rate of abdominal fat thickness compared to before laser irradiation by measuring the fat thickness for 7 times for 90 days using ultrasound after laser irradiation. As a result, fat thickness increased until 1 day after irradiation in all sites. In case of #1, in particular, it increased more than 40 % and then decreased sharply after 7 days. Fat thickness decreased in all areas until 30 days, and then increased by 5 % at #4, #5 and #6 in 60 days. In 90 days, it decreased at all sites. Although there were differences depending on the part, a reduction of 5 % to 35 % in fat thickness was obtained. Irradiation of 1980 nm and 2300 nm combination wavelengths (#1~#5) tended to decrease

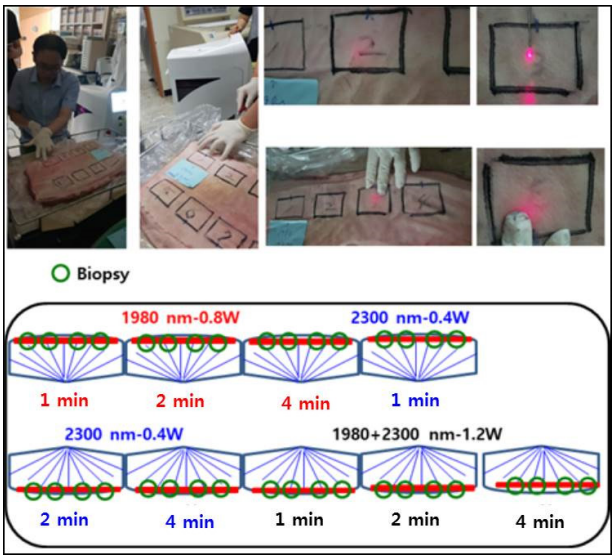

Figure 11. Minimally Invasive Laser Preliminary Experiment Using Pork Bulk Tissue and Laser Irradiation Condition.

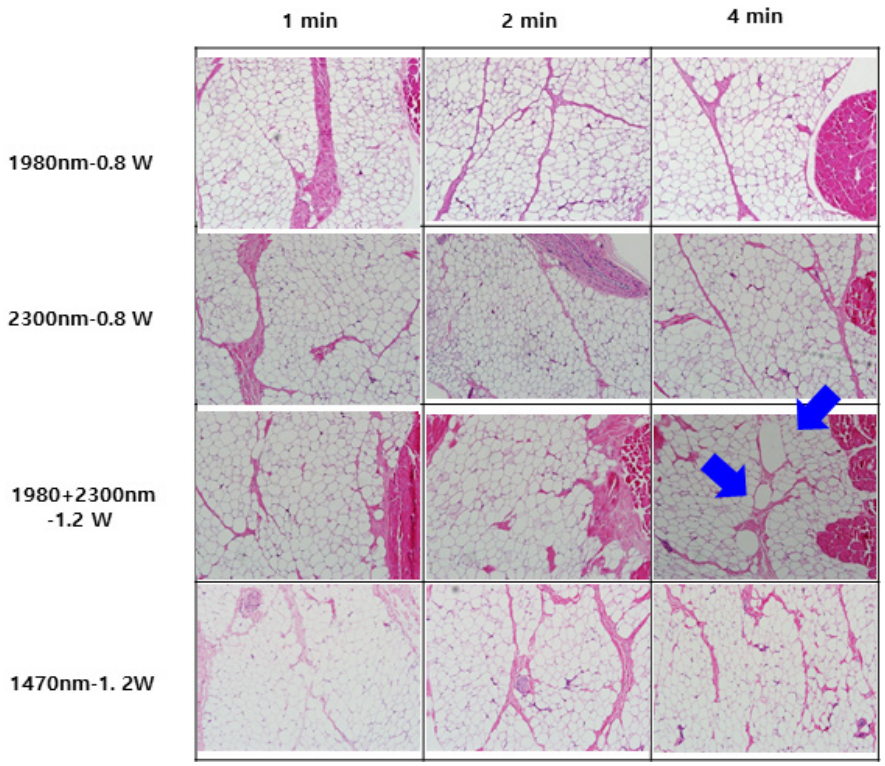

Figure 12. H&E Staining Results of Ex-Vivo Experiments Using Minimally Invasive Laser (×100).

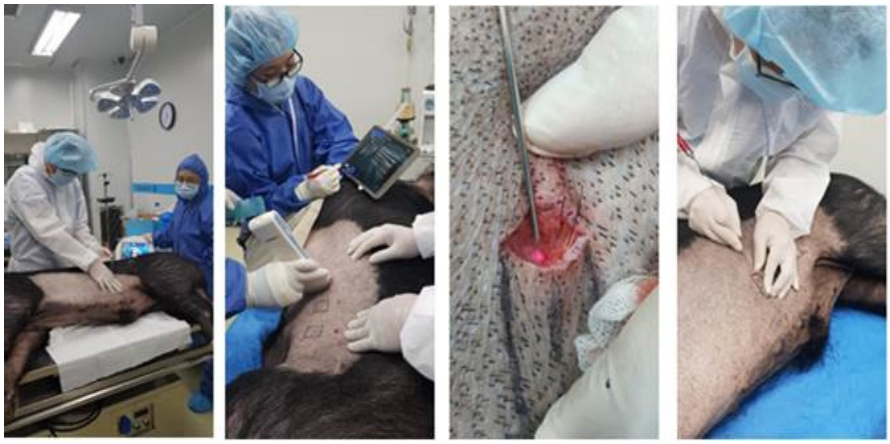

Figure 13. The Process of Minimally Invasive Laser Irradiation on the Fat Layer of Pig Abdomen.

continuously after 60 days. However, in case of #6, which was irradiated with a wavelength of 1470 nm, there was no tendency to decrease after 60 days.

In order to confirm the efficacy and safety in physiopathology, myocytes and fibrous cells were confirmed by H&E staining. Staining resulted in the myocytes and fibrous cells produced between the decomposed adipose tissue in 15 days in the #1, #4, and #5 regions. The 30-day results showed that myocytes and fibrous cells were produced between the adipose tissue decomposed at #2 and #3. The results were similar to those in area #6 (control), but the production of myocytes and fibrous cells was relatively lower than the #1 to #5 areas (experimental group) (Figure 16).

We examined liver function, kidney function, and lipid tests through blood tests. In Table 1, most of the test results did not show a significant change. Although the aspartate aminotransferase (AST) level showed a tendency to increase non-specifically from 4 hours to 24 hours after laser irradiation, it came back to a normal value after 48 hours.

Non-invasive Laser System

We measured the temperature delivered to adipose

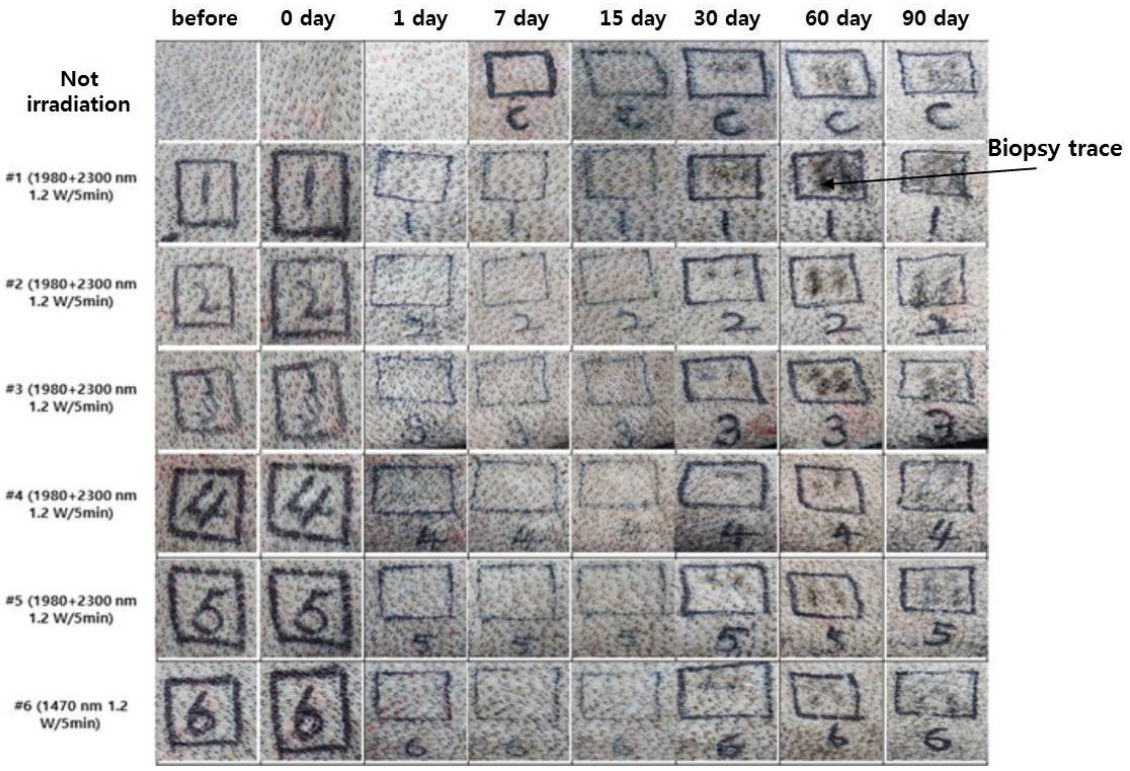

Figure 14. Visual Observation for 90 Days Before and After Minimally Invasive Laser Irradiation.

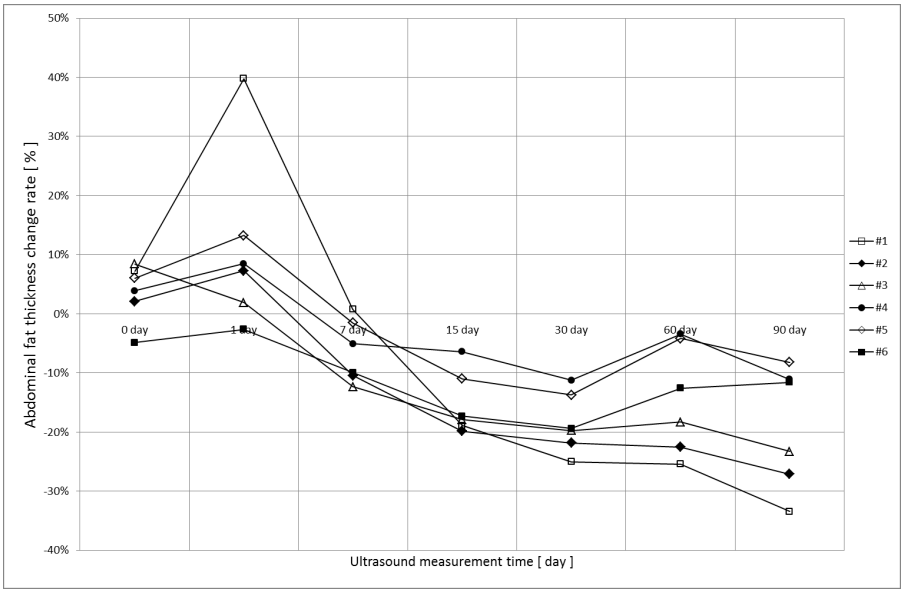

Figure 15. Change in Abdominal Fat by Ultrasound Measurement Before and After Minimally Invasive Laser Irradiation.

tissue during laser irradiation in an ex-vivo experiment. Figure 17 shows the result of measuring the temperature followed by the laser intensity during the 25 minutes of laser irradiation.

As a result of the measurement, after 10 minutes of laser irradiation, the temperature started to rise above 40°C and remained at an average of 48°C until 25 minutes. In the experimental group, three male mini-pigs aged 12 months were used for obtaining multiple results. As shown in Figure 18, each pig abdomen was irradiated with the laser using four hand-pieces. The power of each hand-piece was set differently to 0.8 W/cm^2, 0.9 W/cm^2, 1.0 W/cm^2, and 1.2 W/cm^2. The laser was irradiated for total of 25 minutes. The laser is not irradiated continuously for

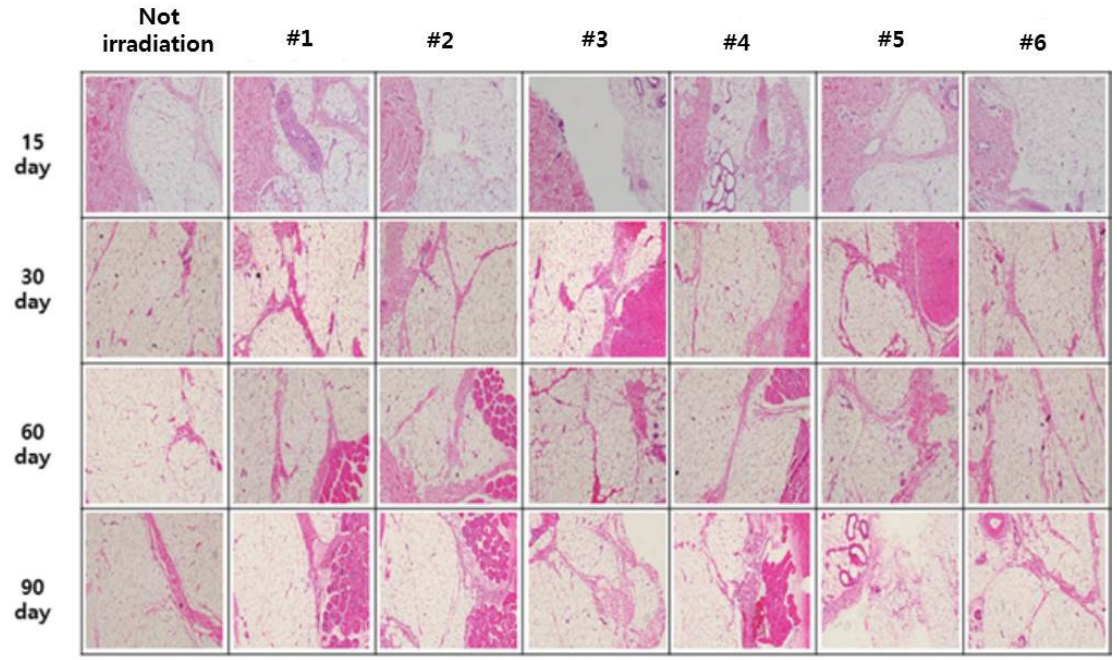

Figure 16. H&E Staining After Minimally Invasive Laser Irradiation (×100).

25 minutes, the laser is irradiated for 20 seconds and 10 seconds break is repeated.

We used a thermal imaging camera to measure the temperature change of the skin before and after laser irradiation. In consequence of the measurement, the average temperature of the pig skin was 33°C (deviation 0.3°C) before irradiation and 30°C (deviation 5.5°C) after irradiation. It indicates that the hand-piece is well cooled. In order to check the safety and effectiveness in this experiment, visual observation, blood test, histology and ultrasound were used to determine the thickness of the fat layer. The biopsy was performed 15, 30, 60 and 90 days after laser irradiation, respectively. Blood tests were performed a total of eight times (0, 2, 4, 8, 12, 18, 24, 48, 72 hours) for 3 days after laser irradiation. Abdominal ultrasound was performed before and after 0, 1, 7, 15, 30, 60 and 90 days respectively. Visual observation showed no significant change in laser irradiation in #1 and #2 pigs. However, as shown in Figure 19, in the case of #3 pig, when the laser intensity was irradiation with 1.2 W/cm^2, curing phenomenon was observed from 15 to 30 days and no curing phenomenon was observed after 60 days.

Figure 20 is a graph showing the average of the change rate of abdominal fat thickness compared to before laser

Table 1. Blood Test Results After Minimally Invasive Laser Irradiation

	TP	ALB	GLB	A/G	AST	ALT	BUN	T-BIL	T-CHO	ALP	Ca	P	TG	CREA	HDL-C	LDL-C	NEFA
	g/dL	g/dL	g/dL	Ratio	IU/L	IU/L	mg/dL	mg/dL	mg/dL	IU/L	mg/dL	mg/dL	mg/dL	mg/dL	mg/dL	mg/dL	mEq/L
D9-15 0H	7.1	4.1	3.0	1.4	47	42	10.4	0.00	88	73	11.1	6.9	23	2.27	40.9	38.9	147
D9-15 2H	6.7	3.9	2.8	1.4	51	39	10.6	0.02	83	75	10.6	6.4	17	2.27	37.9	38.3	113
D9-15 4H	8.1	4.6	3.5	1.3	102	46	14.3	0.05	96	84	11.8	7.4	20	2.54	44.3	43.4	191
D9-15 8H	7.9	4.6	3.3	1.4	160	48	17.8	0.06	90	85	11.4	7.9	19	2.53	41.7	42.1	198
D9-15 12H	7.5	4.4	3.1	1.4	269	48	22.2	0.09	84	86	10.8	8.2	17	2.53	37.1	40.9	198
D9-15 24H	7.3	4.2	3.1	1.4	90	43	19.8	0.02	79	76	10.2	5.4	17	2.22	38.0	35.9	80
D9-15 48H	8.3	4.7	3.6	1.3	57	52	12.4	0.03	101	81	10.9	6.3	21	2.21	48.7	43.7	141
D9-15 72H	7.8	4.5	3.3	1.4	42	52	13.1	0.02	115	86	11.7	5.9	40	2.22	51.1	50.3	36
MIAN	7.6	4.4	3.2	1.4	102	46	15.1	0.04	92	81	11.1	6.8	22	2.35	42.5	41.7	138
SD	0.5	0.3	0.3	0.0	78	5	4.4	0.03	12	5	0.6	1.0	8	0.15	5.2	4.4	59
N	8.0	8.0	8.0	8.0	8	8	8.0	8.00	8	8	8.0	8.0	8	8.00	8.0	8.0	8

TP : Total protein, ALB : Albumin, GLB : Globulin, A/G : Albumin/ Globulin ratio, AST : Aspartate transaminase, ALT : Alanine Aminotransferase, BUN : Blood Urea Nitrogen, T-BIL : Total bilirubin, T-CHO : Total cholesterol, ALP : Alkaline phosphatase, Ca : Calcium, P : Phosphorus, TG : Triglyceride, CREA : Creatinine HDL-C : High density lipoprotein cholesterol, LDL-C : Low density lipoprotein cholesterol, NEFA : Non-esterified fatty acid

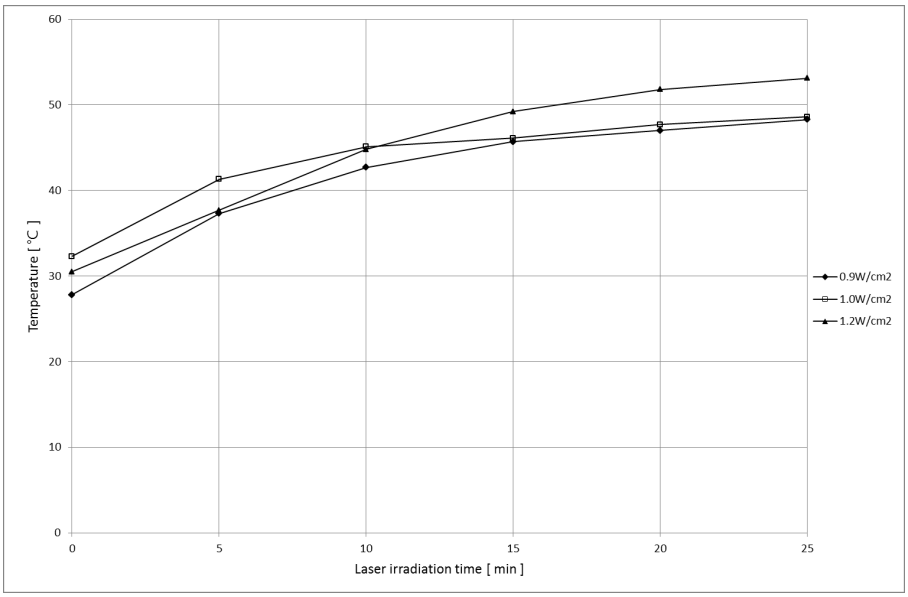

Figure 17. Result of Temperature Change in Pig Tissue by Laser Intensity.

Figure 18. Laser Irradiation Experiments on Pig Abdomen Using a Non-invasive Laser System.

irradiation by measuring the fat thickness of three pigs for 90 days using ultrasound after laser irradiation.

The measurement resulted in a decrease of about 10% except for 1.2 W/cm^2 where curing had occurred after 90 days. In all three pigs, the amount of fat thickness reduction tended to increase as the laser irradiation intensity increased. In addition, it also shows that the change in fat thickness decreases or persists after 90 days. Effective results were obtained with laser irradiation intensity in the range of 1.0 ± 0.1 W/cm.^{2} Figure 21 shows the results of histology. H&E staining showed effective results in all three pigs at the sites irradiated with 1.2 W/cm^2 and 1.0 W/cm^2 of laser intensity after 30 and 60 days. After 90 days, the cellular and fibrous tissues were stably established between adipose tissues.

Particularly, in case of #3 pigs, fibrous tissue was

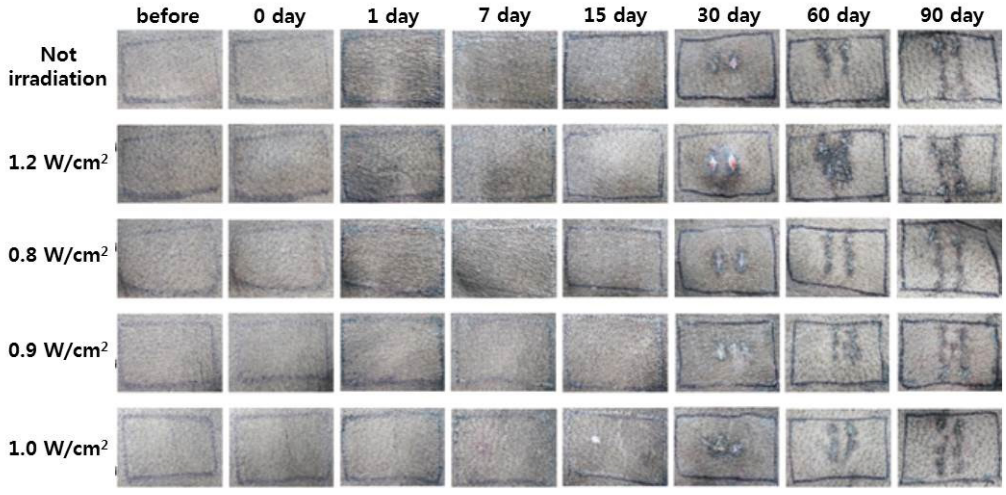

Figure 19. Visual Observation Results for 90 Days Before and After Non-invasive Laser Irradiation in Pig #3.

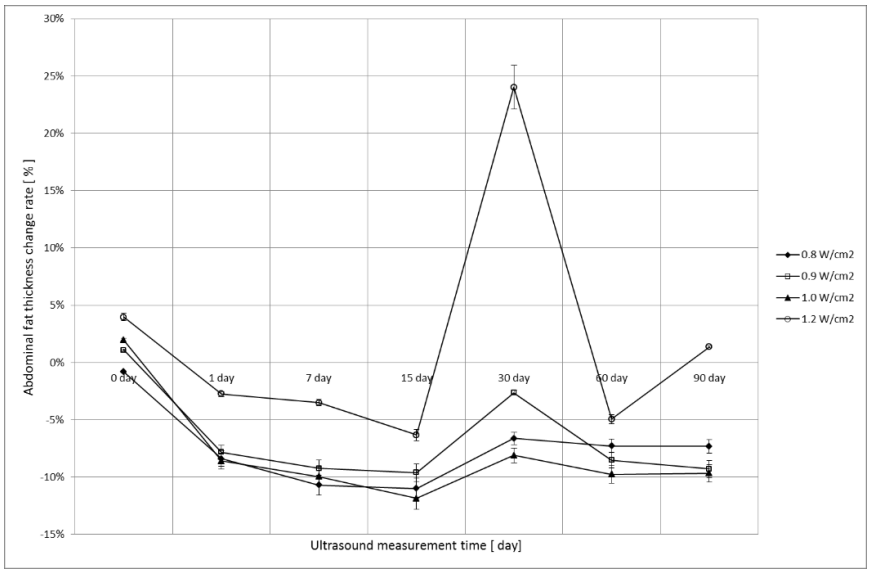

Figure 20. Change in Abdominal Fat by Ultrasound Measurement Before and After Non-invasive Laser Irradiation.

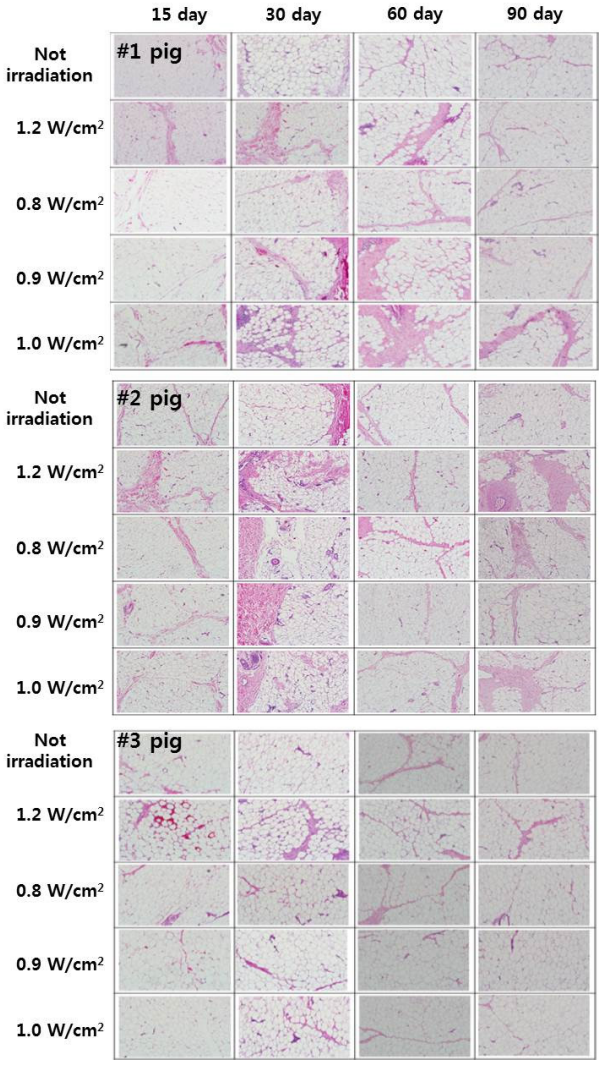

Figure 21. H&E Staining After Non-invasive Laser Irradiation (×100).

filled up between adipose tissue after 15 days when the irradiation intensity was 1.2 W/cm^2. Most of the blood test results in Table 2 showed no significant change. For #3 pig, AST levels increased 11 times in an 8-hour blood sample, but after 12 hours, it was normal. No significant changes were observed in other blood tests.

Combined Minimally Invasive and Non-invasive Laser System

We performed an animal experiment to figure out the lipolysis effect when the minimal invasive laser system and the non-invasive laser system were combined. The experimental group used a female mini-pig, and the pig abdomen was divided into four parts and marked with time points in 0 week, 1 week, 5 weeks and 8 weeks. In parallel experiments, the minimally invasive laser was irradiated 1 day after non-invasive laser irradiation, taking into account the anesthesia time of the pigs.

The laser was irradiated in four conditions as shown in Figure 22 to compare the results. Condition 1 was irradiated with only a 1060 nm wavelength at 1.1 W/cm^2 for 25 minutes with a non-invasive laser system. Condition 2 was irradiated with a wavelength of a 1060 nm for 25 minutes at 1.1 W/cm^2, and the next day, a laser with a wavelength of a 1980 nm was irradiated with a 0.9 W intensity for 5 minutes with a minimally invasive laser system. Condition 3 was irradiated with a wavelength of a 1060 nm for 25 minutes at 1.1 W/cm^2, and the next day, a laser with a wavelength of a 2300 nm was irradiated with a 0.3 W intensity for 5 minutes with a minimally invasive laser system. Condition 4 was irradiated with a wavelength of a 1060 nm for 25 minutes at 1.1 W/cm^2, and the next day, a laser with combination wavelengths of a 1980 nm and 2300 nm was irradiated with a 1.2 W intensity for 5 minutes with a minimally invasive laser

Table 2. Blood Test Results After Non-invasive Laser Irradiation.

	TP	ALB	GLB	A/G	AST	ALT	BUN	T-BIL	T-CHO	ALP	Ca	P	TG	CREA	HDL-C	LDL-C	NEFA
	g/dL	g/dL	g/dL	RATIO	IU/L	IU/L	mg/dL	mg/dL	mg/dL	IU/L	mg/dL	mg/dL	mg/dL	mg/dL	mg/dL	mg/dL	mEq/L
J-16 0H	7.9	4.0	3.9	1.0	40	53	16.5	0.00.	82	55	10.9	5.0	15	2.28	36.4	39.9	14
J-16 2H	8.6	4.1	4.5	0.9	183	70	15.2	0.00	87	58	10.8	5.1	12	2.16	37.5	44.0	53
J-16 4H	8.9	4.2	4.7	0.9	137	69	18.6	0.03	85	61	11.2	6.2	15	2.26	3.6.5	43.5	64
J-16 8H	7.9	3.9	4.0	1.0	106	63	19.7	0.02	77	57	9.9	6.1	20	2.17	32.4	41.9	74
J-16 12H	7.5	3.7	3.8	1.0	70	57	22.4	0.00	74	58	9.3	6.4	28	2.16	29.5	39.3	95
J-16 24H	7.3	3.6	3.7	1.0	75	57	23.6	0.02	71	55	9.9	4.8	21	2.17	28.9	35.9	17
J-16 45H	8.0	4.0	4.0	1.0	67	64	19.5	0.06	86	48	10.4	5.5	18	2.35	31.7	47.8	32
J-16 72H	8.3	4.1	4.2	1.0	61	64	11.6	0.06	99	45	10.2	5.8	32	2.09	35.0	58.8	32
MIAN	8.1	4.0	4.1	1.0	92	62	18.4	0.02	83	55	10.3	5.6	20	2.21	33.5	43.9	48
SD	0.5	0.2	0.3	0.0	47	6	3.9	0.03	9	5	0.6	0.6	7	0.08	3.3	7.0	29
N	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8
J3-32 0H	8.2	4.1	4.1	1.0	23	36	13.1	0.00	70	40	11.2	5.7	16	2.28	37.9	26.3	58
J3-32 2H	9.2	4.4	4.8	0.9	50	40	14.0	0.03	81	45	11.5	5.9	11	2.29	40.9	28.6	140
J3-32 4H	8.5	4.2	4.3	1.0	51	37	16.5	0.02	71	42	10.8	6.0	11	2.32	38.4	27.0	124
J3-32 8H	8.3	4.1	4.2	1.0	54	39	21.0	0.06	68	43	10.8	6.6	11	2.40	36.8	25.0	107
J3-32 12H	8.6	4.3	4.3	1.0	36	39	20.6	0.05	67	45	10.8	5.1	22	2.33	37.9	24.3	61
J3-32 24H	8.2	4.1	4.1	1.0	28	37	19.1	0.03	67	45	10.4	5.4	22	2.27	36.6	23.0	103
J3-32 45H	9.0	4.5	4.5	1.0	29	39	15.3	0.07	80	42	10.9	5.6	14	2.44	41.4	28.8	79
J3-32 72H	8.9	4.5	4.4	1.0	26	41	15.0	0.00	87	46	11.2	5.9	37	2.28	44.0	35.1	11
MIAN	8.6	4.3	4.3	1.0	37	39	16.8	0.03	74	44	11.0	5.8	18	2.33	39.2	27.3	85
SD	0.4	0.2	0.2	0.0	13	2	3.0	0.03	8	2	0.3	0.5	9	0.06	2.6	3.8	42
N	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8
S6-13 0H	7.8	4.7	3.1	1.5	31	50	7.4	0.01	85	57	10.4	5.4	19	1.30	32.3	45.8	47
S6-13 2H	8.7	5.0	3.7	1.4	54	54	8.7	0.00	94	59	11.2	5.6	25	1.35	34.9	519	55
S6-13 4H	8.7	5.0	3.7	1.4	118	64	12.2	0.02	88	56	10.7	5.7	18	1.48	32.4	47.2	67
S6-13 8H	10.4	5.5	4.9	1.1	1312	154	17.5	0.04	87	66	10.7	8.7	29	1.91	31.5	48.1	57
S6-13 12H	8.6	5.1	3.5	1.5	57	57	17.5	0.02	82	68	10.7	6.4	30	1.71	29.2	45.5	147
S6-13 24H	7.8	4.4	3.4	1.3	138	56	11.6	0.00	76	60	10.1	4.9	31	1.26	27.3	41.4	92
S6-13 48H	8.3	4.7	3.6	1.3	61	55	9.7	0.01	99	46	10.2	5.3	23	1.31	31.5	57.9	107
S6-13 72H	7.7	4.5	3.2	1.4	57	50	9.0	0.00	91	43	9.5	5.2	21	1.25	28.9	53.2	102
MIAN	8.5	4.9	3.6	1.4	228.5	67.5	11.7	0.00	87.8	56.9	10.4	5.9	24.5	1.4	31.0	48.9	84.3
SD	0.9	0.4	0.3	0.1	439.3	35.2	3.9	0.00	7.1	8.7	0.5	1.2	5.1	0.2	2.4	5.2	34.0
N	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8

system. We checked the change in the thickness of the fat layer using blood tests, biology and ultrasound to confirm the efficacy and safety. As shown in Figure 23, we observed the rate of change in fat thickness for 8 weeks, which resulted in a minimum of 13 % fat reduction in all conditions.

Fat thickness was confirmed to increase in order of condition 1, condition 3, condition 2, and condition 4. For condition 4, fat thickness was reduced by more than 35 %. Figure 24 shows the results of H&E staining after histopathology. Condition 2, condition 3 and condition 4 irradiated with the laser were confirmed to be fibrous tissue filling up between adipose tissues after 1 week.

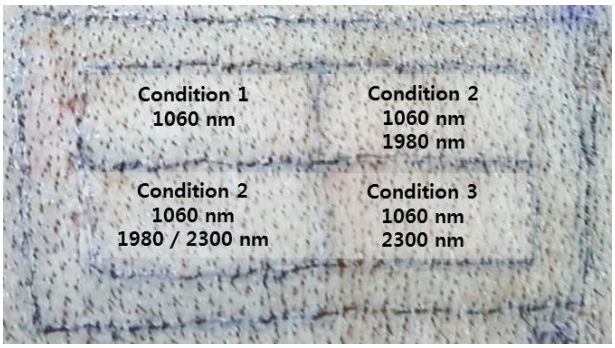

Figure 22. Laser Irradiation Conditions.

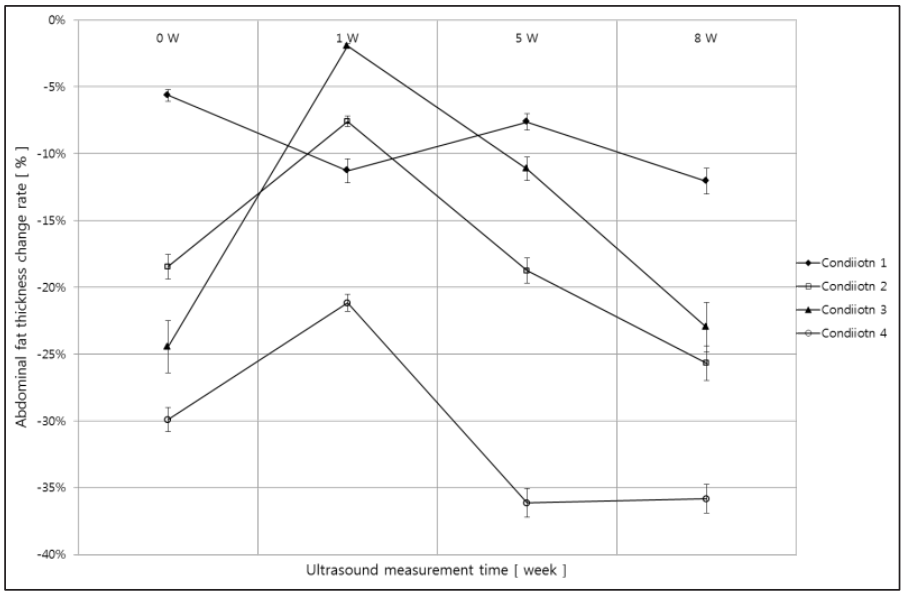

Figure 23. Change in Abdominal fat by Ultrasound Measurement Before and After Combination Laser Irradiation.

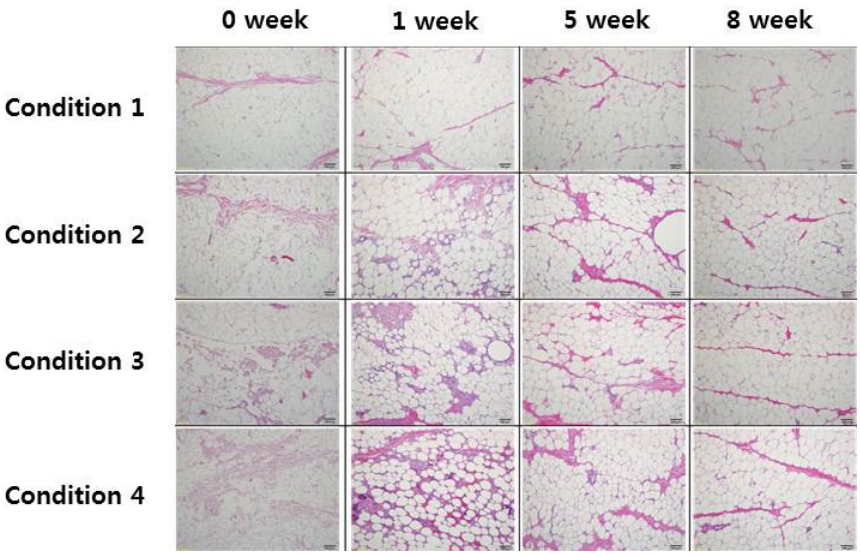

Figure 24. H&E Staining After Combination Laser Irradiation (×100).

After 8 weeks, the fibrous tissue was stably produced between adipose tissues under all conditions.

The blood test was performed 8 weeks after the laser irradiation, and the test items were confirmed to be within the normal range (Table 3).

Discussion

This study aimed to improve the effect of lipolysis by performing a combination of minimally invasive and non-invasive procedures. Related studies are the results of single minimally invasive and non-invasive procedures. For minimally invasive, ACCUSSULPT products have a wavelength of 1444 nm and an output of 12 W.[24] This

Table 3. Blood Test Results After 8 Weeks of Combination Laser Irradiation

	AST	ALT	ALP	TBIL	DBIL	TPRO	ALB	BUN	CREA	TCHO	TG	HDLC	LDLC	IP	CA
	IU/L	IU/L	IU/L	mg/dL	mg/dL	g/dL	g/dL	mg/dL	mg/dL	mg/dL	mg/dL	mg/dL	mg/dL	mg/dL	mg/dL
Value	26.10	31.50	675.12	0.16	0.12	5.93	3.71	11.32	1.40	65.62	40.93	41.39	28.18	7.14	10.82

research uses new wavelengths (1980 nm, 2300 nm) to increase the fat reduction effect with low energy (1.3 W). In the case of non-invasive, the result is the same with the performance as the SCULPSURE product. Regarding laser lipolysis, there is a lot of interest in evaluating the safety and efficacy of clinical trials for most people.[25] The minimally invasive lipolysis laser system using 1980 nm and 2300 nm wavelengths were clinical trials. The non-invasive lipolysis laser system is in the process of obtaining a product approval from the MFDS.

Conclusion

We have developed a minimally invasive lipolysis laser system using non-linear optical crystals to generate wavelengths of 1980 nm and 2300 nm, which have good water and fat absorption. We also developed a non-invasive lipolysis laser system that can be irradiated with four hand-pieces using a 1060 nm diode laser. We performed preclinical studies with pigs to verify the safety and effectiveness of the minimally invasive lipolysis laser system and non-invasive lipolysis laser system. On average, 20% and 6% fat reduction effects were obtained. Blood tests and biopsies confirmed that both systems were safe. In addition, the combination of two systems resulted in a fat reduction of about 35 % and the results of blood tests and biopsies showed no abnormalities in safety.

Ethical Considerations

This study was approved by the research ethics committee of the KBIO HEALTH (Aproval coad #KBIO-IACUC-2018- 064).

Conflict of Interests

None.

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