

***In vitro* fragmentation efficiency of holmium: yttrium-aluminum-garnet (YAG) laser lithotripsy – a comprehensive study encompassing different frequencies, pulse energies, total power levels and laser fibre diameters**

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Objective

To assess the fragmentation (ablation) efficiency of laser lithotripsy along a wide range of pulse energies, frequencies, power settings and different laser fibres, in particular to compare high- with low-frequency lithotripsy using a dynamic and innovative testing procedure free from any human interaction bias.

Materials and Methods

An automated laser fragmentation testing system was developed.

The unmoving laser fibres fired at the surface of an artificial stone while the stone was moved past at a constant velocity, thus creating a fissure.

The lithotripter settings were 0.2–1.2 J pulse energies, 5–40 Hz frequencies, 4–20 W power levels, and 200 and 550 µm core laser fibres.

Fissure width, depth, and volume were analysed and comparisons between laser settings, fibres and ablation rates were made.

Results

Low frequency-high pulse energy (LoFr-HiPE) settings were (up to six times) more ablative than high frequency-low pulse energy (HiFr-LoPE) at the same power levels ($P < 0.001$), as

they produced deeper ($P < 0.01$) and wider ($P < 0.001$) fissures.

There were linear correlations between pulse energy and fragmentation volume, fissure width, and fissure depth (all $P < 0.001$).

Total power did not correlate with fragmentation measurements.

Laser fibre diameter did not affect fragmentation volume ($P = 0.81$), except at very low pulse energies (0.2 J), where the large fibre was less efficient ($P = 0.015$).

Conclusions

At the same total power level, LoFr-HiPE lithotripsy was most efficient. Pulse energy was the key variable that drove fragmentation efficiency.

Attention must be paid to prevent the formation of time-consuming bulky debris and adapt the lithotripter settings to one's needs.

As fibre diameter did not affect fragmentation efficiency, small fibres are preferable due to better scope irrigation and manoeuvrability.

Keywords

holmium:YAG laser lithotripsy, urinary calculi, high frequency lithotripsy, lithotripter settings, automated laser fragmentation testing system

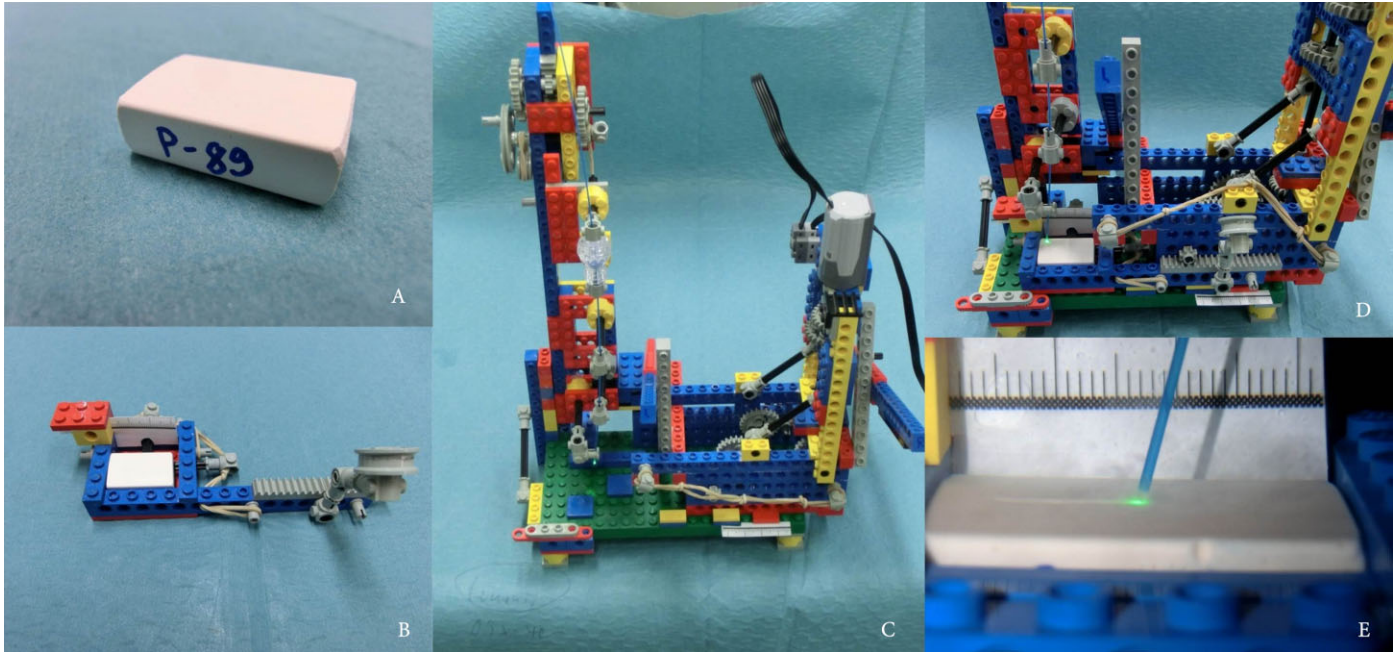
Introduction

The high and rising prevalence of stone disease [1–3] has led to increasing numbers of lithotripsy treatments in urology departments. The opinions of urologists differ about which holmium laser energy setting fragments urinary stones in the quickest and most efficient manner. Some claim that

high-frequency lithotripsy is the most efficient approach [4,5].

To date, the published *in vitro* laser lithotripsy studies have only focused separately on the effect on fragmentation (ablation) efficiency of different laser fibre diameters [6,7], different pulse energies [8,9], or different laser types [10–12].

Fig. 1 Experimental setup and support assembly. **(A)** An artificial stone with a size of $30 \times 20 \times 10$ mm. **(B)** The sliding stone holder bearing an artificial stone. **(C)** The support assembly with the laser fibre holder and the electric motor but without the stone holder. **(D)** The support assembly with the inserted stone holder just before immersion in the tank. **(E)** A close-up of the laser fibre tip and the artificial stone during laser fragmentation in saline. The laser holder maintained the laser fibre at a $75\text{--}80^\circ$ angle (instead of 90°) to prevent it from becoming trapped inside the stone during fragmentation.



A systematic study examining how the various lithotripsy settings interact with each other in shaping fragmentation efficiency has not been performed. In addition, the previous studies all used different experimentation methods. For example, stone samples were subjected to single-pulse laser emission [6,8,13], repeated laser emission [4,14], or just straight-forward perforation [12]. Moreover, in some studies, the majority of tests were performed manually [15–17] and/or used other irregular approaches [14,17,18] that could introduce human bias and/or error.

To address the controversy about the best settings for lithotripsy, the present comprehensive study was performed. Thus, a dynamic and innovative testing procedure that is accurate, yields highly reproducible results, and is free from human interaction was developed. The fragmentation efficiency of high frequency-low pulse energy (HiFr-LoPE) was compared with the fragmentation efficiency of low frequency-high pulse energy (LoFr-HiPE) lithotripsy by measuring fragmentation over wide ranges of pulse energies, pulse frequencies, total power settings, and with different laser fibre diameters.

Materials and Methods

Automated Laser Fragmentation Testing System

To ensure precision, laser fragmentation experiments were performed using an automated system where the immovable

laser fibre fires at the surface of an artificial stone that is moved along an axis at a constant velocity, thereby engraving a fissure on the stone surface. The stone was chosen to be the moving component instead of the laser fibre as this meant a less complicated automated mechanism could be used, thus ensuring accuracy.

The system consisted of an automated experimentation support assembly that was made from Lego Technic™ (Fig. 1). The assembly included a sliding holder that kept the artificial stone in place while it was moved at a constant velocity (21 mm/30 s) along a horizontal plane by an electric motor for 30 s (during which time the stone moved a distance of exactly 21 mm). The assembly also included a specially designed holder for the laser fibre that kept the fibre tip at $75\text{--}80^\circ$ and allowed the tip to move slightly and adapt itself to changes in the stone surface that arose from stone fragmentation. This ensured that the fibre tip always remained in contact with the stone material. It also prevented the fibre tip from becoming trapped in a crater and being dragged away by the sliding stone. The whole support assembly was immersed in a tank filled with 0.9% saline.

Preparation of Artificial Stones

Artificial stones were made from plaster of Paris, using the appropriate powder to water ratio of 1.5:1 by weight [19]. The stones were left to dry for >96 h at room temperature. The

Fig. 2 Surface photographs of the artificial stones that were exposed to high- or low-frequency laser emissions from laser fibres whose diameters were 200 or 550 μm and which were used at power levels that ranged from 5 to 12 W. Naked eye observations show that low-frequency emissions clearly had superior outcomes. This was confirmed by subsequent digital volumetric measurements. The large 550 μm fibre produces wide but shallow fissures, while the small 200 μm fibre generates deep but narrow fissures.

Power Level (W)		High Frequency	Laser Fibre (μm)	Low Frequency	
5	25 Hz x 0.2 J		200		10 Hz x 0.5 J
			550		
6	30 Hz x 0.2 J		200		5 Hz x 1.2 J
			550		
8	40 Hz x 0.2 J		200		10 Hz x 0.8 J
			550		
12	40 Hz x 0.3 J		200		10 Hz x 1.2 J
			550		

tensile strength of these stones is very similar to several types of urinary stones [20]. Moreover, they are composed of a relatively soft material [21]. This makes them suitable for measuring fragmentation at very low pulse energies that might not yield detectable fragmentation if harder materials were used.

Before the experiment, the artificial stones were submerged in 0.9% saline and soaked in a uniform manner. The experiment began when the stone holder was put into motion, laser emission was initiated, and a fissure was carved into the stone.

Lithotripsy Fibres

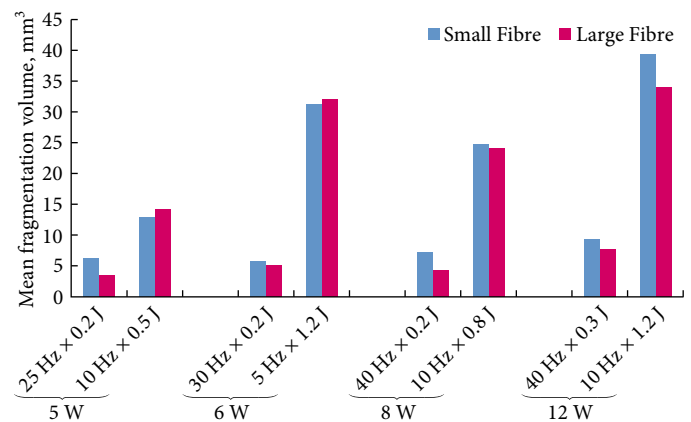
To assess possible variations resulting from differently sized laser fibres, each experiment was performed with either a small 200 or large 550 μm core laser fibre (Lumenis – SlimLine™). Brand new laser fibres were used and the tip was cleaved after each experiment to avoid any possible fibre tip degradation bias.

Lithotripsy Settings

A broad range of laser lithotripter settings was used. There were three groups of experiments. The high- vs low-frequency experiment involved 16 different settings with total power levels of 5, 6, 8 and 12 W, pulse energies of 0.2, 0.3, 0.5, 0.8, and 1.2 J, pulse frequencies of 5, 10, 25, 30, and 40 Hz, and small and large fibres. The combinations are specified in Figs 2 and 3.

The 6 W experiments, which were performed to more closely evaluate the changes in fragmentation efficiency as frequency declined and pulse energy rose, included four experiments of the previous test group and eight additional experiments at pulse energies of 0.2, 0.3, 0.4, 0.6, 1.0, and 1.2 J, and pulse frequencies of 5, 6, 10, 15, 20 and 30 Hz. The combinations, which were performed with small and large fibres, are specified in Table 1.

Fig. 3 The mean fragmentation volume achieved by small (200 μm) and large (550 μm) fibres at four different power levels and at high-or low-frequency lithotripter settings.



Another 10 experiments involved the following five W/Hz/J combinations, which were performed with both small and large fibres: 4/8/0.5, 6.4/8/0.8, 9.6/8/1.2, 16/40/0.4, and 20/40/0.5. These 10 experiments were added to the 16 high- vs low-frequency experiments and the eight additional 6 W experiments to make the 34 test dataset displayed and analysed in Fig. 4.

The laser lithotripter used was the Lumenis™ VersaPulse® Powersuite 100 W, which can be used at these multiple frequency and pulse energy settings.

Fissure Measurements

After the experiments were completed, the stones were dried for >96 h at room temperature and the fissure width, depth, and volume were measured. Fissure depth was measured

Table 1 Relationship between fragmentation and pulse energy at the 6 W power level. Comparison of the fragmentation (ablation) rates with the initial fragmentation volumes († and ‡) reveals an almost directly proportional increase as pulse energy rises for both types of laser fibres. If the pulse energy level doubled, tripled or even quintupled in relation to the initial high frequency setting, the fragmented volume ratios rose in an equivalent fashion although the power level remained the same (6 W).

Frequency, Hz	Pulse energy, J	Small 200 µm fibre		Large 550 µm fibre		Proportional increase of initial pulse energy (0.2 J)
		Mean (sd) FV, mm ³	Ratio of FV to initial FV [†]	Mean (sd) FV, mm ³	Ratio of FV to initial FV [‡]	
30	0.2	5.73 [†] (0.05)	1.0	5.18 [‡] (0.18)	1.0	1.0
20	0.3	8.57 (0.07)	1.5	11.32 (0.46)	2.2	1.5
15	0.4	10.67 (0.21)	1.9	12.27 (0.71)	2.4	2.0
10	0.6	17.59 (1.08)	3.1	21.34 (1.59)	4.1	3.0
6	1.0	24.46 (1.65)	4.3	25.73 (1.72)	5.0	5.0
5	1.2	31.30 (0.78)	5.5	31.98 (0.75)	6.2	6.0

FV, fragmentation volume.

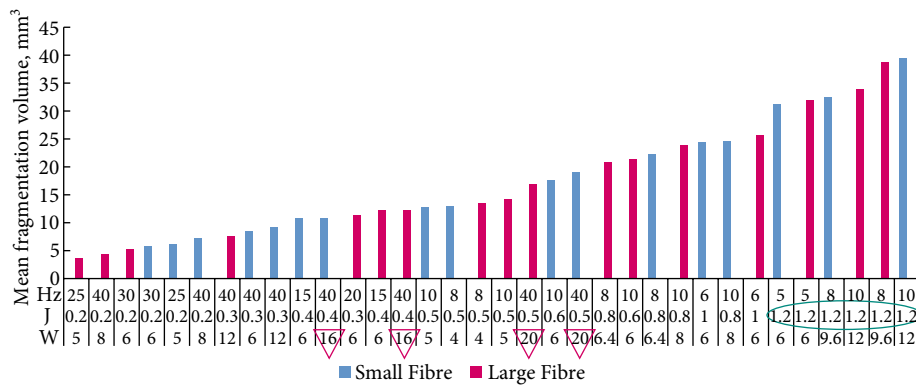


Fig. 4 Organisation of the 34 experiment dataset according to mean fragmentation volume. High frequencies at higher power settings (inverted red triangles) did not yield higher fragmentation rates than LoFr-HiPE settings, even when the settings of the latter were several orders of magnitude smaller. By contrast, the highest pulse energies were associated with the highest fragmentation volumes (green circle).

accurately by using the depth of focus of a calibrated optical microscope (Nikon Labophot 2) [22]. Concerning fissure width, the borders of the fissure were sometimes too irregular to make exact measurements. To overcome this, high resolution photographs were taken with tangential lighting to reveal the outlines of the fissure. After photographic calibration and use of image processing software (ImageJ 1.46r, U.S. National Institutes of Health, Bethesda, Maryland, USA), the depth, width, and volume of the fissure were calculated. The volume measurement served as the primary measure of fragmentation efficiency. Fissure width and depth served as secondary measures.

Statistical Analyses

Statistical analysis was performed using StatEL 2.6, AD Science, Paris, France. The *t*-test was used to compare high- vs low-frequency settings and small vs large laser fibres. Pearson's correlation test was used to evaluate relationships between the different laser settings and the resulting ablation rates. A *P* < 0.05 was considered to indicate statistical significance.

Results

Figures 2 and 3 show the high- vs low-frequency experimental data. At the same power level, LoFr-HiPE lithotripsy achieved

a higher fragmentation (ablation) volume than HiFr-LoPE for all power levels that were tested, regardless of the fibre size. Compared with HiFr-LoPE, LoFr-HiPE increased the volume on average by 4.5 (SD 1.4)-fold (*P* < 0.001).

When the mean fragmented volumes of the 16 LoFr-HiPE vs HiFr-LoPE tests were ordered from the lowest to the highest volumes, the tests with the highest pulse energy (1.2 J) congregated at the higher end. When these tests were combined with another 18 tests involving other power, pulse energy, and pulse frequency combinations (see above), thus yielding 34 tests, the same distribution was seen (Fig. 4). There was a linear correlation between pulse energy and fragmentation volume (*P* < 0.001). However, the power (W) used did not correlate with fragmentation volume (*P* = 0.29).

The results of secondary experiments at the 6 W power level are shown in Table 1. This indicated that gradual increases in pulse energy were accompanied by steady, almost proportional, increases in mean fragmented volume. Thus, compared with the initial high-frequency setting, increasing the pulse energy essentially increased the fragmented volume ratios in a proportional manner.

Analyses of fissure width and depth revealed that these variables showed similar relationships as the mean fragmentation volume. First, LoFr-HiPE settings were

associated with significantly wider and deeper fissures than HiFr-LoPE ($P < 0.001$ and $P < 0.01$, respectively). Second, analysis of the 34 experiments revealed that higher pulse energy correlated significantly with wider ($P < 0.001$) and deeper fissures ($P < 0.001$).

When different laser fibres were used for these experiments, statistically significant fragmentation volume changes were not detected. This was generally true even when the experiments were grouped according to regular total power or pulse energy intervals ($P = 0.81$). The exception was at very low pulse energies: at the energy of 0.2 J, the 550- μm fibre was significantly less effective at fragmentation than the smaller 200- μm fibre ($P = 0.015$). Notably, large fibres were always associated with wide fissures ($P < 0.001$) and small fibres were always associated with deep fissures ($P < 0.001$), regardless of the frequency, pulse energy, or total power used.

It was observed during the experiments that the high-frequency experiments generated very small, almost powder-like fragments, whereas low-frequency lithotripsy generated slightly larger fragments. However, all of these debris pieces were very friable and turned completely into dust. Consequently, measurable fragments were not available for further evaluation.

Discussion

There are only a few studies on high-frequency laser lithotripsy. One evaluated the effect of pulse frequency and the so-called 'popcorn effect' on multiple loose stone fragments. It was found that while increasing frequency tended to increase the fragmentation rate, there was a decline when very high frequency and higher energy settings were used [4]. The present study showed that at the same power levels, HiFr-LoPE was significantly less efficient than LoFr-HiPE lithotripsy.

The present study also showed that the fragmentation (ablation) volume, fissure width, and fissure depth increased when the pulse energy rose. This relationship between fragmentation volume and pulse energy has already been described in the early publications of leading experts in the field, such as Teichman and Vassar [8,9]. However, these single-pulse laser emission studies analysed the effect of pulse energy on its own; the possible interactive effects of different pulse frequency or total power were not examined. A more recent study that included high-frequency tests found that when the same frequency is maintained, fragmentation increases four-fold when 10-times as much pulse energy is applied [17]. In the present study, the effect on fragmentation efficiency of varying both frequency and total power levels was examined. This revealed that higher total power settings did not guarantee higher fragmentation volumes or wider or deeper fissures, even when significantly higher total power was used compared to LoFr-HiPE (Fig. 4). The secondary

experiments at the 6 W power level also revealed another interesting association (Table 1): as the pulse energy increased, the mean fragmented volume ratio increased in an almost directly proportional way. In other words, if the pulse energy level doubled, tripled or even quintupled in relation to the initial high-frequency setting, the fragmented volume ratios rose in an equivalent fashion. This indicates that the key variable that affects fragmentation efficiency is the pulse energy, whereas frequency, especially high frequency, and total power setting play more minor roles.

Several studies have shown that low pulse energy settings produce smaller stone fragments than high pulse energy settings [5,23]. This was also marginally observed in the present study, although no measurable fragments resulting from any of the lithotripter settings tested were recovered. Many endourologists claim that HiFr-LoPE settings assist in achieving a more thorough stone fragmentation because they 'dust' the stone and leave no fragments to remove afterwards; nevertheless the present study shows that these same settings can be up to six-times slower if one considers the total ablation volume. But one admits that *in vivo* stone material (other than plaster of Paris) may possibly produce some bulky debris with LoFr-HiPE, debris whose removal may be time-consuming. The influence of debris size and the importance of uniformly dusting the stone should not be underestimated, as indicated by the following example of three different-sized stones whose length is 1, 2 or 3 cm, respectively. These stones have a stone volume of 1, 8, and 27 mL, respectively, and their fragmentation into 5-mm fragments would result in 8, 64 or 216 5-mm fragments, respectively. All of these fragments would have to be removed individually. If a 10/12 F ureteric access-sheath was used, the stones would have to be reduced to 3-mm fragments, which is the maximum stone size that can be accommodated by this sheath. In the case of the 3-cm stone, 1000 3-mm fragments would be produced. During laser lithotripsy, whether using the slower HiFr-LoPE setting or the faster LoFr-HiPE setting, it is important to ensure that the lithotripter setting that is applied ablates the stone uniformly and does not generate numerous large fragments. The removal of such fragments significantly increases the operating time, may require the use of a stone-removal device or specific laser lithotripsy techniques (e.g. the 'popcorn effect') to reduce the stone burden [4]. Cutting of the stone into small, removable pieces should be considered only when a small residual stone volume remains. Thus, slower ablating HiFr-LoPE, but only dust producing settings can be a useful element in the fragmentation procedure as a whole. Similar studies using real-life stone material are needed to confirm these results.

The studies comparing large and small laser fibres have yielded contradictory results: some suggest that at the same laser lithotripter settings, large fibres fragment large volumes [13], whereas others have found the opposite [9]. In the

present study, large fibres always created wider fissures, while small fibres always generated deep fissures, but neither surpassed the other in terms of fragmentation volume. Indeed, for any given laser setting (with the exception of very low energy settings), the small fibre produced narrower yet deeper fissures than the large fibre. As a result, the two fibres fragmented similar volumes. The disparities between the present study and those of others may relate to the experimental approaches that were used. When isolated single laser pulses [8,9,13] or repeated laser pulses scattered over a surface are used [14,17], or handheld experiments are conducted [15,17], the impact of the laser never or rarely occurs at the same location twice. If the stone structure around these isolated fragmentation craters is weakened by the first impact, any new impacts on the same site could generate a significantly higher fragmentation volume. However, this effect cannot be detected by the above-described study designs. By contrast, the present study used continuous laser emission, where the same place was hit several times while moving the fibre steadily along a path. This closely mimics real-life lithotripsy and allows the effect of repeated laser emission impacts on the same area to be measured. This may explain the differences between the present and previous studies.

The large and small laser fibres differed only at the smallest energy level that was tested, namely, at 0.2 J when the large fibre was less effective. This may relate to energy dispersion. As the tip of the large laser fibre has a significantly larger contact area (7.5-times bigger) than the small fibre, the amount of energy delivered at the 0.2 J level may be too dispersed to exceed the ablation threshold [11].

Although the present study showed that the laser fibre diameters that were tested did not influence the laser ablation rate directly, the physical properties of the fibres can still exert a positive or negative effect on the lithotripsy procedure overall. Large-diameter laser fibres are more resistant to fibre tip degradation or the 'burn-back' effect, but their stiffness hampers scope deflection and their diameter obstructs irrigation flow. By contrast, small-diameter laser fibres are more fragile and prone to degradation phenomena with higher energies, but facilitate ureterorenoscopy flexibility and irrigation rates at the same irrigation pressure, thereby increasing accessibility and visibility [7,9,14,24,25]. As fragmentation volume is not affected, the advantages of small-diameter laser fibres, namely, their increased scope deflection facility and irrigation rate (and consequent better visibility), lead us to prefer and recommend the use of small-diameter laser fibres, notwithstanding the financial costs associated with increased fibre degradation.

The present study did not take retropulsion into account, and did so intentionally. Retropulsion is affected by known factors, e.g. stone mass and pulse energy [11,13,26], the latter being

one of the main variables analysed, but retropulsion is probably also influenced by other features like stone shape, friction between the surrounding tissue and the stone, etc. With the exception of pulse energy, all the previous variables could introduce potentially bias, leading to irreproducible results and conceal otherwise significant outcomes concerning pulse energy, frequency, total power and laser fibre diameter, the real focus of the present study. Nevertheless, retropulsion poses a problem to urologists because it reduces the laser's efficiency, increases operating time, and sometimes makes the calculus inaccessible. Some of the recommendations the present study advises might be impractical for very small free floating stones (e.g. minor ureteric calculi), because retropulsion increases with rising pulse energy [11,13]. However, in those stones only slightly influenced or completely unaffected by retropulsion due to size, location (e.g. trapped in a calyx), being fixed or impacted, our recommendations of using LoFr-HiPE can be useful and accelerate the procedure, especially on the brink of changing guidelines considering constantly larger stones suitable for retrograde intrarenal surgery (RIRS) [27,28].

The present study showed that should urologists wish to use the same total power level, they can accelerate their lithotripsy procedure simply by increasing the pulse energy and reducing frequency, although attention must be paid to preventing the production of bulky debris that might have an opposing effect and prolong the procedure. It also showed that urologists can use small-diameter laser fibres without compromising stone ablation efficiency, with the added benefit of having better irrigation, visibility, and manoeuvrability. Conventional laser lithotripters continue to offer as much as other high-frequency lithotripters at the power levels tested without jeopardising the fragmentation rate or surgery time.

The limitations of the present study concern the recommendation's applicability to very small calculi and the associated retropulsion. In the future, the next experimental step would be to mimic laser lithotripsy of very small non-impacted ureteric calculi. However, to include the simultaneous analysis of lithotripter settings, ablation volume and retropulsion of such small stones, and examine how they influence each other using this experimental setup, one would have to develop a system, where, besides all regular variables used, the retropulsion effect needs to be measurable and independent but still somehow 'mechanised' and consistent, regardless of the different experimental parameters used. We are not sure if such a model can be developed that integrates ablation rate and retropulsion in a mechanised and reproducible fashion without introducing new factors, influencing existing ones, and leaving others to chance. This dilemma may be similar in analogy to Heisenberg's uncertainty principle in particle physics: one may either know the precise velocity of a particle or know its exact location, but never both of them at the same time [29].

The experimental setup of the present study met all planned criteria and proposed objectives that were described in the Introduction and Materials and Methods sections. Strenuous efforts were made to limit possible bias. Future comparisons with other testing methods will assess the power and reliability of the present study.

Acknowledgments

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Conflict of Interest

None declared.

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Abbreviations: HiFr-LoPE, high frequency-low pulse energy; LoFr-HiPE, low frequency-high pulse energy.