CUSTOM DESIGNED ACOUSTIC PULSES

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(Paper JBO-199 received Apr. 22, 1998; revised manuscript received Aug. 19, 1998; accepted for publication Sep. 18, 1998.)

ABSTRACT
We have used a tunable, infrared, free-electron laser with a Pockels cell controlled pulse duration to generate photoacoustic pulses with separate variable rise times (from 15 to 100 ns), durations (100–400 ns), and amplitudes (0.005–0.1 MPa). The tunability of the free-electron laser across water absorption bands allows the rise time of the thermal-elastically generated acoustical pulse to be varied, while a Pockels cell controls the duration and cross polarizers control the pressure amplitude. The mechanical effects of pressure transients on biological tissue can have dramatic consequences. In addition to cell necrosis, carefully controlled pressure transients can also be used for therapeutic applications, such as drug delivery and gene therapy. This technique permits systemic probing of how biological tissue is affected by stress transients. © 1999 Society of Photo-Optical Instrumentation Engineers. [S1083-3668(99)00302-0]

Keywords photoacoustic liquids; free electron lasers; biological acoustics; stress waves; computer simulations.

1 INTRODUCTION

It is desirable to understand the biological effects of the laser-induced pressure transients. There are many mechanisms that produce laser-induced pressure transients: optical breakdown, ablation, thermal expansion, electrostriction, and radiation pressure. These mechanisms are extensively reviewed in the literature (see, for example, Ref. 1). The dominant mechanism for acoustic wave generation at lower fluences is thermal expansion. Thermoelastic stress waves are produced by the thermal expansion of the sample after absorption of the laser energy. The exact form of the thermoelastic wave depends on the geometry of the source and sample, the laser temporal and spatial profiles, and the thermal and acoustic properties of the sample. For a more extensive description of thermoelastic stress waves see, for example, Ref. 2.

Recently it has been discovered that pressure waves transiently open the cell membrane, temporarily increasing the permeability of the cell membrane.3–6 This effect can be used to enhance the intracellular concentration of different drugs or allow the delivery of drugs that are normally impermeable to the cell membrane while maintaining cell viability.7 Larger amplitude stress waves can kill cells directly. Several measurements have shown that shock waves can suppress tumor growth.8 Experiments by Doukas et al.9 have shown that cell survival decreases with increasing pressure but is also dependent upon the rise time of the pressure transient. When the fraction of cells that survived was plotted versus the stress gradient (peak pressure divided by the rise time), the data collected at different rise times coalesced, implying that the stress gradient is biologically important. There are indications that the increase in permeability of the cell membrane is also sensitive to the stress gradient (unpublished data).

Although progress has been made in understanding the interaction of pressure transients with tissue, the complete biological effects of pressure transients are not completely understood. One of the main hindrances to the investigation of the biological effects of stress transients has been the inability to control the shape of the stress pulse. In this article, we present a method of controlling the acoustic pulse shape by using the thermoelastic mechanism and pulses from a free-electron laser (FEL).

2 METHOD

2.1 THEORY

For the current discussion we will consider stress waves generated in a fluid, although the one-dimensional solution of the wave equation would be the same for tissue. Provided the rate of thermal expansion is small compared to the speed of sound, the linearized hydrodynamic equations for a homogeneous fluid can be used. If the influence of heat conduction within the sample is negligible on the time scale of the measurements, and nonlinear ef-
fecteds and attenuation can be ignored, the following
inhomogeneous wave equation can be derived: 10
\[ \nabla^2 P(r,t) - (1/c_s^2) \partial^2 P(r,t)/\partial t^2 = -\left(\beta/C_p\right) Q/\partial t, \]  
(1)

where \( P \) is the pressure, \( c_s \) is the sound velocity, \( \beta \)
is the coefficient of thermal expansion, \( C_p \) is the
specific heat at constant pressure, and \( Q \) is the heat
generated from the absorbed laser light per unit
volume per unit time. For the experiments reported
here, the beam diameter, \( d \), is much greater than the
absorption depth of the sample, \( \mu_a^{-1} \). Under these
conditions, the boundary between the near field
(planar waves) and far field (spherical waves) zones, \( z_f \), is given by\(^{11} \)

\[ z_f = d^2/4\lambda_{ac}, \]  
(2)

where the acoustic wavelength (\( \lambda_{ac} \)) is determined by the
absorption depth or the laser pulse duration and the speed of sound depending on whether the
absorption depth is greater than or less than the
distance of acoustic propagation during the laser
pulse. Our sample thickness was always shorter
than \( z_f \), allowing the generated acoustic pulses to be
treated as planar. The general one-dimensional
solution to Eq. (1) for \( d/2 > \mu_a^{-1} \) is given by Burmis-
trova et al.\(^{12} \) For the work presented here, a rigid
boundary was applied to create a unipolar pulse.
For a rigid boundary and a spatially uniform laser beam profile,

\[ P(\tau) = C_s (c_s t_0 / 2\pi)^{1/2} \int_{-\infty}^{\infty} f(t')K_0(\tau-t')dt', \]  
(3)

where

\[ K_0(\tau-t') = (\pi A^2/t_L)\exp(-A|\tau-t'|/t_L) \]  
(4)
is the transfer function of the thermo-optical con-
version layer, \( \tau = t - z/c_s \) is the reduced time, \( t_L \) is the
laser pulse width, \( A = \mu_a c_s t_L, I_0 \) is the laser
peak irradiance, and \( f(t) \) is the temporal pulse
shape. Although the above equation was derived for a spatially uniform laser beam, the same result is obtained for a Gaussian beam profile (TEM\(_{00}\) mode).\(^{13} \)

In the case where the laser pulse is short com-
pared to the time necessary for an acoustic pulse to
propagate a distance equal to the absorption depth,
\( 1/(\mu_a c_s) \), the resulting pressure wave becomes in-
dependent of the laser pulse shape. At distances much larger than the absorption depth (\( z \gg \mu_a^{-1} \)),
the pressure is given by

\[ P(\tau) = \epsilon_s \beta I_0 / (2C_p) \times \]
\[ \left\{ \begin{array}{ll}
1 - \exp(-\mu_a c_s t_L) & \mu_s<\tau, \\
2 - \exp(-\mu_a c_s \tau) - \exp(-\mu_s c_s (\tau-t_L)) & 0 < \tau < t_L, \\
\exp(-\mu_s c_s t_L) - 1 & \tau - t_L \end{array} \right\} \]  
(5)

In the opposite regime, where the laser pulse is long compared to \( (\mu_a c_s)^{-1} \), \( A \gg 1 \) and \( K_0(\tau-t') \)
approximates a delta function multiplied by a factor of \( 2\pi A \). Thus, the acoustic pulse shape follows the
profile of the laser pulse,

\[ P(\tau) = (c_s \beta I_0 / C_p) \tau. \]  
(6)

The pulse structure of the Vanderbilt University
FEL consists of a 5 \( \mu \)s macropulse composed of sub-
picosecond micropulses spaced 350 ps apart. The
duration of the micropulse (\( \leqslant 1 \) ps) is negligible on
acoustic time scales. Provided the absorbed energy is
rapidly converted into heat, the pressure transient
produced by a single micropulse is given by Eq. (5).
Since \( t_L \ll (\mu_a c_s)^{-1} \), we can approximate the
pressure profile at depths \( z \gg \mu_a^{-1} \) as

\[ P_{\mu}(z,t) = P_0 \exp(-\mu_a(z-c_s t)), \]  
(7)

where \( P_0 \), the peak pressure, is given by

\[ P_0 = \mu_a c_s^2 \beta I_0 t_L / (2C_v). \]  
(8)

From Eqs. (7) and (8) it can be seen that the acoustic
wave form generated by a micropulse depends upon
the optical absorption coefficient and the sound velocity. Since the peak pressure depends on
the absorption coefficient, both the rise time and the
amplitude will change with laser wavelength.
However, the rise time is independent of laser irradi-
ance so any change in the peak pressure that occurs
from varying the laser wavelength can be com-
penbated for by changing the laser irradiance. The
amplitude of the acoustic pulse also depends upon
the coefficient of thermal expansion and the heat
capacity. We use the heat capacity at constant vol-
ume, \( C_v \), with the short laser pulses. These param-
ters are temperature dependent and the shape and
amplitude of the acoustic pulses may change as the
optical pulse heats the sample. Provided these tem-
perature dependent changes are small, the pressure
wave generated by the overall optical pulse can be
approximated by a superposition of the pressure
transients generated by the individual micropulses:

\[ P_M(z,t) = \sum_{i=1}^{N} I_i P_{\mu}(z,t-(i-1)t_m), \]  
(9)
\[ P_M(z,t) = \sum_{i=1}^{N} I_i P_0 \exp(-\mu_d(\lambda)) \times \left| z - c_5[(t-(i-1)t_m)] \right|, \]

where \( I_i \) is the irradiance of the \( i \)th micropulse relative to the peak irradiance \( I_0 \), \( t_m \) is the time between micropulses, and \( N \) is the number of micropulses in the optical pulse.

### 2.2 EXPERIMENT

Experiments were performed using the Vanderbilt FEL with a modified output coupler. To control the optical pulse duration, a Pockels cell was designed with a 15 ns rise time and an adjustable duration between 100 and 1000 ns. The timing of the pulse (truncated with the Pockels cell) relative to the leading edge of the macropulse was adjustable, allowing the transmitted region of the macropulse to be selected. A schematic of the experimental setup is shown in Figure 1. The TEM\(_{00}\) output of the FEL was put through an antireflection coated silicon or germanium window to remove the harmonics that would either pass through the Pockels cell or potentially damage the cadmium telluride (CdTe) crystal. The FEL beam was reduced to 3 mm in diameter with a BaF\(_2\) lens telescope and passed through two zinc selenide (ZnSe) Brewster plate polarizers that were used to adjust the laser irradiance. The beam was then passed through a CdTe Pockels cell and an additional ZnSe Brewster plate polarizer. The Pockels cell described by Becker et al.\(^{14}\) was used for collecting data at 6.6 \( \mu \)m (EOM3-UC-0347-C-BC, II–IV). All other data were collected with a new Pockels cell (EOM-5-2.0-5.0-0540-C, II–IV) which had the following modifications: (1) the crystal faces were perpendicular to the laser beam rather than cut at a Brewster angle and coated with a broad band antireflection coating, and (2) the electronics were reversed so that the laser beam was transmitted only when the high voltage was applied. The extinction ratio of the Pockels cell was at least 150:1. The pulse duration was measured by reflecting a small portion of the laser beam with a BaF\(_2\) window to a photoelectromagnetic (PEM) diode (PEM-L, BSA Technology). The signal was recorded without amplification by a digital storage scope (HP54522A, Hewlett Packard). The sample was mounted on a 9 \( \mu \)m thick polyvinylidene fluoride (PVDF) piezoelectric detector. The propagation distance from the window to the detector was determined by a spacer. A sapphire window was placed over the sample (water for these experiments) and fastened in place by a top plate to provide a rigid boundary. The PVDF detector was coupled to a Plexiglas base for impedance matching and the output was recorded by a transient digitizer (7912 HB, Tektronix). A unity gain buffer was used to isolate the detector from the input impedance of the digitizer. The beam diameter at the sample was 3 mm.

The acoustic detector was calibrated by applying a well defined ballistic pressure (a metal ball was dropped from measured heights). For the 3 mm beam diameter used in our experiments, the sensitivity of the detector was 0.1 bar/mV. The temporal resolution of the transducer and unity gain buffer was tested by measuring the stress wave generated by plasma formation when irradiating aluminum foil with 35 ps pulses from a Nd:YAG laser. The resolution of the system was found to be \( \sim 6 \) ns.

All experiments reported here were performed on water. The water was purified by a Milli-Q (Millipore, Bedford, MA) filter system. Absorption from the O–H stretch band of water changes by more than two orders of magnitude between 2.94 and 3.8 \( \mu \)m,\(^{15}\) making it possible to produce stress waves with rise times that range from nanoseconds to hundreds of nanoseconds. Experiments were performed at 3.21 \( \mu \)m (\( \mu_a = 0.338 \mu \text{m}^{-1} \)), 3.30 \( \mu \)m (\( \mu_a = 0.140 \mu \text{m}^{-1} \)), 3.78 \( \mu \)m (\( \mu_a = 0.011 \mu \text{m}^{-1} \)), and 6.60 \( \mu \)m (\( \mu_a = 0.068 \mu \text{m}^{-1} \)). Typical pressures in these experiments ranged from 1/10 to 1 bar. The laser irradiances used for all experiments were believed to be subthreshold for cavitation and other nonlinear effects.

### 3 RESULTS

Figure 2(a) shows a comparison of single acoustic pulses generated at 3.3 \( \mu \)m with laser pulse durations of 100, 200, 300, and 400 ns at a power of \( \sim 6 \) kW during the laser pulse (calculated from 30 mJ of...
energy for a 5 μs long pulse). Since we are interested in the pulse shape, the data set was normalized to the maximum pressure of the 400 ns pulses, but the relative amplitudes were unchanged. The corresponding laser profile measured with the PEM diode is shown in Figure 2(b). The rise time and amplitude of the acoustic pulses are independent of duration while the duration of the acoustic pulses follows the duration of the laser pulse. The acoustic pulse begins to form a tensile component with increasing pulse duration. This effect is attributed to diffraction since it increases with acoustic wavelength and sample thickness. We note that the optical pulses show a tail that increases with increasing duration. This phenomenon occurs in the CdTe Pockels cell around 3 μm and is dependent on laser irradiance and alignment. However, the optical pulse tail is not responsible for the tensile component of the acoustic pulse since the tensile component is observable in data taken at 6.6 μm where the optical tail is absent.

Figure 3 shows single acoustic pulses generated by a 100 ns laser pulse at 3.3 μm (μcCL_L = 20.8) and 3.78 μm (μcCL_L = 1.6) with a pulse power of 6 kW (30 mJ from the 5 μs pulse) and 1 kW (5 mJ from the 5 μs pulse), respectively. The duration and amplitude of the pulses are similar, but the rise and decay times are markedly different. Experiments were performed at 3.21 μm with different laser irradiances (Figure 4). The acoustic pulse shape remained unchanged as the laser pulse power was varied from 0.3 to 1 kW (1.7–5 mJ from the 5 μs pulse).

Figure 5(a) shows results from experiments at 3.3 μm for a 100 ns laser pulse with an average power of 6 kW (30 mJ from the 5 μs pulse) measured at depths of 0.8 and 1.6 mm along with the calculated rise time based on Eq. (10). Data and theory for experiments at 3.78 μm for a 100 ns, 1 kW average power (5 mJ from 5 μs pulse) laser pulse are shown in Figure 5(b).

4 DISCUSSION

The calculated rise time from Eq. (10) using an ideal square pulse for the macropulse envelope is shown in Figure 5(a). The calculated rise time is faster than what is experimentally observed. By using the actual optical pulse to determine I1 in the calculation, the discrepancy was decreased. The remaining difference is most probably caused by the temporal resolution of the detector. The calculations suggest...
that the 10 ns rise time of our optical pulse affects the rise time of the generated acoustic pulse. Using a variable rise and decay time for the optical pulse and the appropriate selection of absorption coefficient, it is possible to experimentally vary the rise time and decay time independently. The Pockels cell electronics can be designed to give a sharp rise time within an adjustable fall time. Then the rise time will be dependent upon the absorption coefficient of the sample while the decay time will follow the envelope of the optical pulse. The reverse scenario is also possible where the rise time depends upon the optical pulse envelope and the decay time is determined by the absorption coefficient of the sample. Thus, all four parameters (rise time, duration, decay time, and amplitude) can be varied independently.

Figure 5(b) shows a comparison between experiment and theory for an acoustic pulse generated by a 100 ns laser pulse at 3.78 μm with 6 kW pulse power (30 mJ from the 5 μs pulse). Here, the acoustic rise time is considerably slower than the rise time of the optical pulse and there is good agreement between experiment and Eq. (10) using an ideal square pulse for the optical pulse envelope. The data taken at 6.6 μm are similar to the data at 3.78 μm and are not shown here. The results at 6.6 μm signify that the generated acoustic pulse is dependent on the absorption coefficient, but not on the resonance absorbing the laser pulse.

The rise time of the curves in Figure 5(a) shows no significant broadening or steepening and the amplitude shows no significant attenuation between the measured depths of 0.8 and 1.6 mm. Therefore, we feel justified in ignoring the effects of attenuation and nonlinear processes for these measurements. An increase in the tensile component occurs at larger propagation depths, supporting the claim that diffraction is responsible for the formation of the tensile component.

The pulse structure of the FEL is well suited for the generation of custom acoustic pulses. The 350 ps separation of the micropulses delays the onset of nonlinear effects by temporally distributing the laser energy. The micropulses are spaced close enough together that the acoustic pulses from the individual micropulses are not distinguishable. Pressure transients of several hundred ns duration can be created and fit well to the linear superposition of individual acoustic pulses. It is also possible to find a portion of the FEL macropulse that is relatively flat, keeping the laser irradiance constant during creation of the pressure transient.

Figures 2–4 verify that the rise time, duration, and peak amplitude of the generated acoustic pulses can be varied independently. Other methods are available for generating stress waves with variable forms such as laser ablation and the propagation distance through media. Zweig et al. found that the rise times of pressure transients generated by ablating polyamide varied with the laser wavelength. However, they also saw that the rise time changed with the laser irradiance. The dependence of the rise time on the fluence prohibits one from using this method to make a systematic investigation of the biological effects. Allowing the acoustic pulse to propagate through a medium before impinging on the sample can also be used to alter the characteristics of the pressure transient. The attributes of the stress wave depend upon the linear and nonlinear properties of the medium. In general, the sound and particle velocity increases with increasing stress, causing the leading edge of the stress wave to steepen and form a shock wave. The linear acoustic properties of the medium will attenuate the high frequency components of the acoustic pulse. Therefore, the rise time of the stress wave can, in principle, be controlled by choosing the correct propagation distance in a medium with the appropriate acoustic properties. The drawbacks to this approach are that it is cumbersome and the overall amplitude is attenuated.

In the initial studies reported here, the amplitude of the acoustic pulses generated was relatively low. Typical pressures ranged between 1/10 and 1 bar, well below the peak pressures of 0.1–1 kbar cur-

![Figure 5](https://www.spiedigitallibrary.org/journals/Journal-of-Biomedical-Optics/figure5.jpg)
rently used to investigate biological effects. However, the peak pressure can be raised considerably by increasing the laser power, by decreasing the laser spot size, or by using a sample with a larger Grünenisen coefficient. Walsh and Cummings have shown that the optical properties of water change at high laser power. This imposes a limit on what laser irradiances can be used without having to consider nonlinear effects. Vodopyanov measured 100 J/cm³ as the threshold for photobleaching in water and 100 GW/cm² for saturation of the O–H stretching mode as measured with HDO in D₂O.¹⁹ The maximum absorbed energy per unit volume for our experiments was 12 J/cm³ and the maximum irradiance of the individual micropulses was approximately 30 MW/cm². Nonlinear effects must be considered when using higher irradiance pulses. These nonlinear effects can include the nonlinear response of the tissue to the laser light, the diffraction effects from the nonplanar waves, and the creation of cavitation bubbles. Cavitation was observed for pulse durations longer than 500 ns when the beam diameter was decreased to ~0.5 mm. The peak pressures generated were in excess of 30 bar. Cavitation can be avoided by decreasing the duration of the laser pulse. The ability to generate short stress waves with fast rise times is an advantage of the thermoelastic mechanism. An acoustic pulse of 10 ns duration in tissue corresponds to a spatial width of 15 μm. As the spatial rise time and duration decrease, the entire stress pulse will occur over the scale of a single cell and the stress gradient over an even smaller distance. Subnanosecond stress pulses have already been generated in liquids.²⁰ It will be interesting to see what novel effects occur when ultrashort stress waves are applied to tissue. It has been shown that the stress gradient is an important parameter in cellular damage. With fast Pockels cell electronics, acoustic rise times of 1 ns are achievable. The peak pressure one needs to obtain the same stress gradient decreases as the rise time becomes faster, making it possible to study the biological effects of stress waves using lower peak pressures.

5 CONCLUDING REMARKS

The pressure waves generated by the interaction of lasers and tissue have been considered for the collateral damage that they can cause in biological tissue. However, recently there has been a growing interest in the possible therapeutic applications of these pressure transients. Progress towards clinical applications has been hampered by lack of a detailed understanding of the effects of the characteristic features of pressure transients on tissue. To study these effects, it is necessary to develop methods for controlling the pressure transient parameters independently. In this article we have presented methods for varying the rise time, duration, and peak pressure of the pressure transients. The rise time is controlled by the absorption depth of the sample that can be varied by changing the wavelength of the laser. The duration is varied using a Pockels cell while leaving the amplitude and rise time unchanged. Cross polarizers alter the pressure amplitude without changing the pulse shape. The macropulse structure of the FEL is ideal for creating custom designed acoustic pulses. The pressures transients generated by the individual micropulses can be summed together to give the shape of the stress wave generated by the whole laser pulse. The 350 ps separation between micropulses extends the pulses durations that one can achieve before nonlinear effects become important. These methods should allow investigators to perform more systematic evaluations of the effects of pressure transients in biological (as well as other) systems.

Acknowledgments

The authors would like to thank Klaus Becker for his help with the Pockels cell electronics. This work was supported by the Department of Defense Medical Free Electron Laser Program under Grant Nos. N00014-87-C-0146 (Vanderbilt University), N00014-91-C-0084, and N00014-94-I-0927 (Wellman Laboratories of Photomedicine).

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