

Reducing Radiation Damage in Soft Matter with Femtosecond Timed Single-Electron Packets

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10 **Significance Statement:** Specimen damage is a major limiting factor in electron microscopy. A
number of approaches exist for mitigating negative effects, but challenges remain. Advances in
pulsed electron sources are intriguing, but little is known about the effects on damage,
particularly when compared to conventional methods. Here, using a femtosecond pulsed laser to
control electron emission and deliver each electron to the specimen in a regular, precisely-timed
15 manner, the authors show that damage can be reduced compared to conventional approaches for
the same dose rate and total dose. Further, damage is shown to depend on the time between
arrival of each electron and on the number of electrons arriving in a given amount of time,
indicating specimen relaxation reduces the damage incurred.

Abstract: Despite development of myriad mitigation methods, radiation damage continues to be a major limiting factor in transmission electron microscopy. Intriguing results have been reported using pulsed-laser driven and chopped electron beams for modulated dose delivery, but the underlying relationships and effects remain unclear. Indeed, delivering precisely-timed single-electron packets to the specimen has yet to be systematically explored, and no direct comparisons to conventional methods within a common parameter space have been made. Here, using a model linear saturated hydrocarbon (*n*-hexatriacontane, C₃₆H₇₄), we show that precisely-timed delivery of each electron to the specimen, with a well-defined and uniform time between arrival, leads to a repeatable reduction in damage compared to conventional ultralow-dose methods for the same dose rate and the same accumulated dose. Using a femtosecond pulsed laser to confine the probability of electron emission to a 300-fs temporal window, we find damage to be sensitively dependent on the time between electron arrival (controlled with the laser repetition rate) and on the number of electrons per packet (controlled with the laser-pulse energy). Relative arrival times of 5, 20, and 100 μs were tested for electron packets comprised of, on average, 1, 5, and 20 electrons. In general, damage increased with decreasing time between electrons and with increasing electron number. Further, we find that improvements relative to conventional methods vanish once a threshold number of electrons per packet is reached. The results indicate that enhanced specimen recovery enabled by precise electron-by-electron dose delivery leads to a repeatable reduction in irreversible structural damage.

In transmission electron microscopy (TEM), structural damage caused during electron-specimen interactions results in significant degradation of spatial resolution, which ultimately limits the quality and the accuracy of the information obtained. Indeed, the progression of damage during exposure leads to uncertainties in agreement between the acquired data and the intrinsic (*i.e.*, artifact-free) properties of the material. The specific nature of the damage and the overall degree to which it occurs depends on a number of variables; including the electron-beam characteristics, the specimen properties, and the experimental requirements (1). As such, development of methods for minimizing deleterious effects of beam damage and for providing routes to elucidating the nature and the relative influence of fundamental mechanisms at work requires detailed, systematic exploration of a large, multivariable parameter space.

Though the factors and the mechanisms associated with beam damage are numerous and multi-faceted, a number of practical approaches have been shown to significantly reduce negative effects (2). For example, cryogenic methods operate by holding the specimen at reduced temperatures during exposure in order to lower diffusion and kinetic reaction rates, to reduce mass loss, and to minimize the impact of beam-induced thermal effects, especially for poorly-conducting materials (3, 4). This has led to significant advances in the ability to study highly-sensitive specimens, such as soft matter and biological structures, at sub-nanometer spatial scales (5-8). Minimizing exposure of the specimen to electron irradiation *via* low-dose and low-dose-rate methods and the use of conductive coatings and substrates have also proven beneficial (9-13). Further related to controlling damage through dose, the method of dose fractionation, which is based on the acquisition of fast, multi-frame scans or brief, stroboscopic multi-image series, has been shown to be a viable approach for minimizing beam-induced artifacts (14-16).

Transfer of energy from an incident electron to a material initiates a cascade of structural and electronic excitations, each of which is active within a characteristic temporal window (17). Accordingly, interesting questions can be posed pertaining to the feasibility of taking advantage of relaxation and self-healing processes in order to mitigate damage. Indeed, self-healing has been implicated in the recovery and the dose-rate-dependent damage thresholds observed for inorganic materials, biomolecules, and tissues (18-21). In light of this, delivering dose in a segmented fashion with a well-defined duration between each packet of electrons is a potentially intriguing approach to reducing or even circumventing the negative effects of certain damage mechanisms. Interestingly, the effects of pulsed-electron irradiation on damage to tissues, cells, and biomolecules is studied in the field of radiobiology, often by utilizing femtosecond (fs) pulsed-laser plasma accelerators (21, 22). In electron microscopy, the potential benefits of using a pulsed electron beam to mitigate damage have been speculated upon for decades (23-25). Further, though not attempting to leverage relaxation and self-healing, proposals for extending the concept of diffract-before-destroy to TEM have also been discussed (26-29). In this approach, sufficient information would be acquired before any damage has occurred by using a single, brief, large number-density electron packet. Unlike X-ray photons, however, electrons in such a dense packet will experience significant Coulomb repulsion, which will degrade coherence and limit resolutions (30, 31). This unwanted effect necessitates the use of high energies and/or post-emission recompression schemes (32-35).

In order to systematically test the effects on damage of using a pulsed source in the TEM, and especially for direct comparisons to conventional methods, one ideally would have an extremely high level of control over key aspects of the discrete electron packets. Such aspects include the number of electrons in each packet, the duration of each packet, the time between

each packet, and the number of packets per unit time. As will be described in more detail below, implementation of fs laser-driven photocathodes and picosecond chopped beams in TEMs provides such control (24, 25, 36, 37), and a few intriguing observations noting potential damage reduction when using a pulsed source have indeed been reported. For example, diffraction-spot intensities from crystals of copper 7,7,8,8-tetracyanoquinodimethane and polyethylene oxide under pulsed-beam illumination from a laser-driven photocathode were observed to be stable over inordinately long continuous exposure times (38, 39). More recently, apparent mitigation of damage was observed in MgCl_2 when using a segmented beam consisting of picosecond electron bunches, each regularly spaced in time by ~ 160 ps (*i.e.*, a repetition rate of 6 GHz) (40). However, as in pulsed-beam radiobiology studies (41), the precise effects on damage of pulsing the beam, and the relationship with dose rate, are largely unknown. Indeed, no study has yet determined the fundamental effects of precisely delivering *each individual electron* to the specimen with an extremely well-defined and highly-tunable time between each. Further, under such finely-controlled conditions, no direct comparisons have been made to conventional low-dose methods *for precisely the same dose rate and the same total dose*.

Here, we report our observations of a repeatable reduction in damage when using pulsed electron packets generated *via* fs laser excitation of a photocathode in a modified TEM as compared to a conventional thermionic beam for precisely the same dose rate and accumulated dose. By using fs laser excitation to precisely control the electron-emission process, we confine the probability of emission to a temporal range of 300 fs (full-width at half-maximum, FWHM) for extremely regular peak-to-peak emission times. Using a Bragg-spot fading-curve method and a model beam-sensitive soft-matter material (*n*-hexatriacontane, $\text{C}_{36}\text{H}_{74}$, C36 hereafter), we measure a nearly two-fold reduction in damage when the dose is delivered *one electron at a time*

with up to 100 μs between the arrival of each at the specimen. That is, for thermionic beams of the same dose rate [*e.g.*, as low as $7.8 \times 10^{-7} \text{ e} \cdot \text{\AA}^{-2} \cdot \text{s}^{-1}$; 1.6 femtoamp (fA) beam current; 0.01 zeptoamps per nm^2], the observed damage is nearly double that of the pulsed beam at a common total dose. We also explore the effects of varying the duration between the arrival of each individual electron at the specimen and the number of electrons in each individual packet. In general, we find that reducing the duration between electron arrival or increasing the number of electrons in each packet leads to an increase in damage to the point where the effects of pulsing the beam are indistinguishable from conventional approaches, indicative of a threshold effect.

Results and Discussion

Pulsed-Beam Method. An overview of the concept and approach is provided in Fig. 1. The key advance leveraged here is the use of a fs laser to control electron emission from the TEM source (see Fig. S1 in the Supporting Information). In this way, one can dictate precisely when emission occurs, how often it occurs, and how many electrons are emitted during each pulse. Here, emission is confined to a temporal window as small as 300 fs FWHM (laser pulse duration), is restricted to occurring as infrequently as once every 100 μs (laser repetition rate, f), and is limited to as few as one electron per pulse [laser pulse energy; see Fig. S2]. As a result, a precisely-defined, extremely-regular specimen rest time can be applied by varying f . Regularity arises from the temporal emission probability [$P(t)$] being confined to the laser-pulse duration; in pulsed mode, emission occurs only when photons are incident on the photocathode source [*i.e.*, $P(t) = 0$ between laser pulses]. As such, electron emission is restricted to occurring within a temporal window defined as $f^{-1} \pm 150 \text{ fs}$ for a train of 300-fs laser pulses (for negligible laser pulse-to-pulse jitter). Note that subsequent electron acceleration and mismatches in photon

energy and cathode work function will add an effective timing jitter approximately on the order of the laser-pulse duration (42). During a particular experiment, shot-to-shot pulse-energy stability and statistics of photoemission dictate the number of electrons emitted during each pulse. Here, the stability was such that an error of approximately $\pm 10\%$ (one standard deviation from the mean) was measured for one electron per packet, while it was substantially smaller for 5 and 20 electrons per packet (see Fig. S2 and Table S1). Note that the value of $P(t)$ for conventional thermionic emission is both non-zero and constant during the entire exposure time (*i.e.*, emission probability is the same throughout). It is also important to note that long-term stability of the pulsed photoelectron beam was high, varying for example by $\pm 0.3\%$ over a 24-hour period during a successful experiment [see Fig. S3].

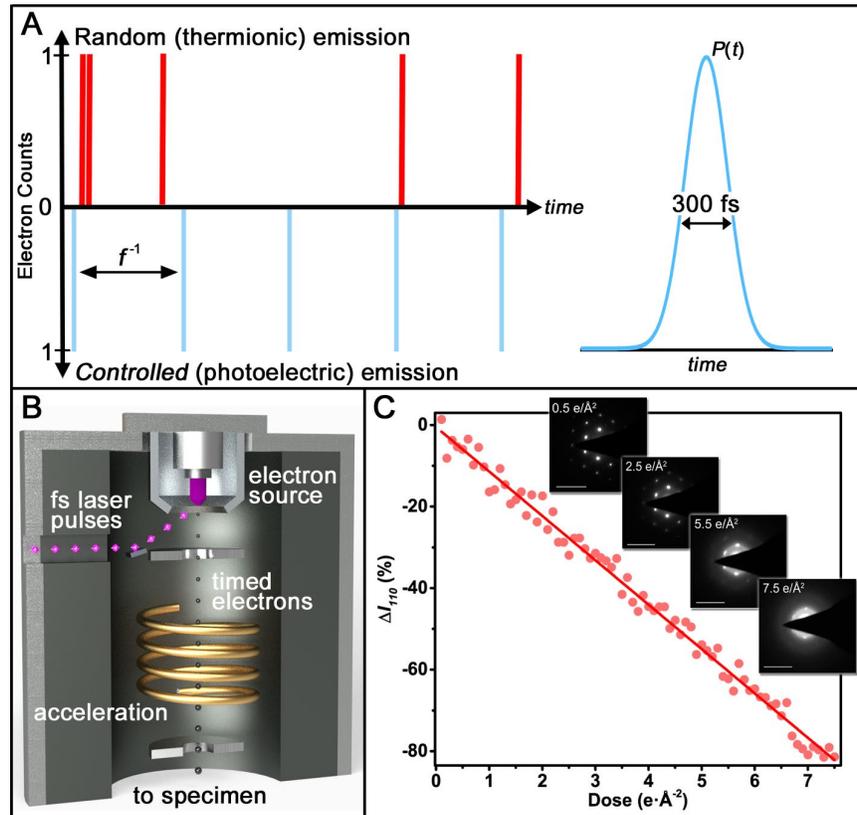


Fig. 1. Experimental approach. (A) Concept of controlled, precisely-timed electron emission compared to conventional random (thermionic) emission for the same beam current. The laser repetition rate (f ; pulses per second) dictates the time between electron emission (f^{-1}) for the pulsed approach. Laser-pulse energy is tuned to select the desired number of electrons, and the temporal emission probability [$P(t)$] is confined to the pulse duration (here, 300 fs FWHM). (B) Schematic of the source region of the modified TEM. The fs laser pulses enter through an optical port and are directed onto the source to generate timed electron packets. These are then accelerated and directed to the specimen using conventional methods. (C) Example of the Bragg-spot fading-curve method used to monitor damage [see also Fig. S4]. Shown is representative data of the fading of 110 Bragg spots from a C36 microcrystal during random (thermionic) irradiation. Here, $\Delta I_{110} = (I_{110} - I_{o,110})/I_{o,110}$, where I_{110} is the average intensity of the 110 spots at a specific dose ($\text{e}\cdot\text{\AA}^{-2}$), and $I_{o,110}$ is the initial average intensity at nominally zero dose.

One Electron Per Packet, 100 μs Between Each Electron. Effects of pulsed- and random thermionic-beam irradiation on 110 Bragg-spot intensities from individual C36 microcrystals for an identical ultralow dose rate of $7.8 \times 10^{-7} \text{ e}\cdot\text{\AA}^{-2}\cdot\text{s}^{-1}$ are summarized in Fig. 2 (see the Supporting Information for a description of the specimen preparation). For the pulsed mode, the number of electrons generated per laser pulse was 1.03 ± 0.15 , and the time between arrival of each at the C36 microcrystal was $f^{-1} = 100 \mu\text{s}$ (see the Supporting Information for details on the measurement of the number of electrons per packet). Intensities were monitored up to an accumulated dose of $0.066 \text{ e}\cdot\text{\AA}^{-2}$ (see also Fig. S5). Linear intensity decreases were observed for each over this range. When elastic-bending and dynamical-scattering effects are negligible,

Bragg-spot intensity attenuation as a function of dose is indicative of a loss of crystallinity due to increasing disorder and irreversible structural damage (43). Here, the slopes of the responses differ by a factor of 1.8, with the rate of damage sustained for the pulsed beam being lower than that of the thermionic beam despite being applied at the same dose rate. See Figs. S6 and S7 and the Supporting Information for descriptions of the methods used to measure beam current and beam size. Note that care was taken to ensure the C36 microcrystals studied were the same thickness [see Fig. S8 and the Supporting Information].

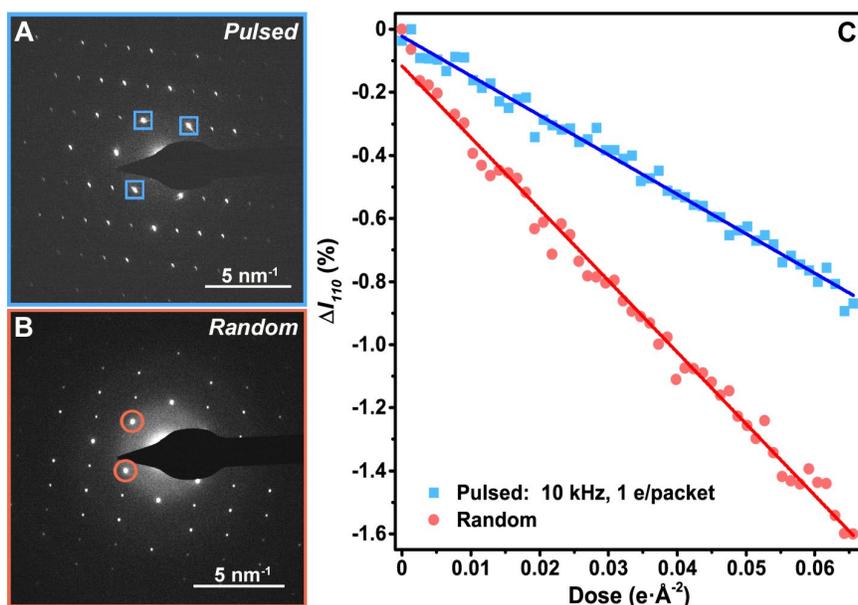


Fig. 2. Comparison of pulsed and random (thermionic) electron-beam damage for the same dose rate ($7.8 \times 10^{-7} \text{ e} \cdot \text{\AA}^{-2} \cdot \text{s}^{-1}$). Representative (A) pulsed- and (B) random-beam diffraction patterns of a C36 microcrystal with the 110 Bragg spots used to generate the fading curves in panel (C) highlighted. (C) Fading curves and least-squares fits for the pulsed and random beams. The slope of the line fit to the random-beam data is steeper than that of the pulsed beam by a factor of 1.8. Pertinent specifications are as follows: Pulsed-beam current and illuminated area were $1.6 \pm 0.24 \text{ fA}$ and $132.4 \text{ }\mu\text{m}^2$, respectively ($0.01 \text{ zeptoamps per nm}^2$). Random-beam current and

illuminated area were 1.6 ± 0.22 fA and $131.5 \mu\text{m}^2$, respectively. The pulsed beam was generated with a 300-fs FWHM pulsed laser operated at $f = 10$ kHz ($f^{-1} = 100 \mu\text{s}$). The laser pulse energy incident on the TEM photocathode was set such that the number of electrons per packet was measured to be 1.03 ± 0.15 .

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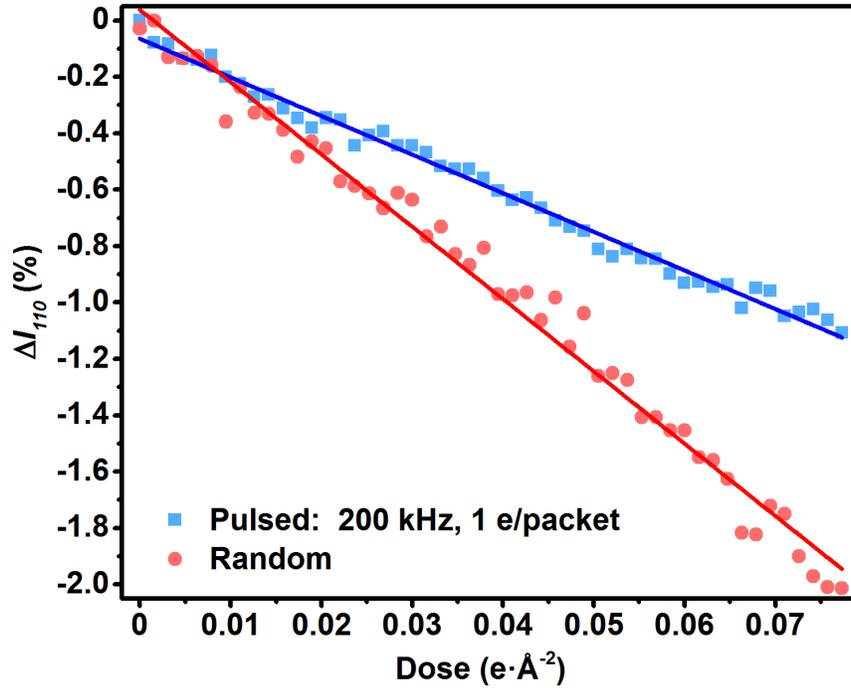
The results shown in Fig. 2 suggest that relaxation and recovery from damage mechanisms that are potentially reversible on timescales shorter than f^{-1} (*e.g.*, $100 \mu\text{s}$) occurs to a significantly greater extent than with conventional low-dose methods, at least at the ultralow doses and ultralow dose rates applied here. In insulators like C36, such mechanisms include
10 electron transitions and secondary-electron generation, phonon excitation and specimen heating, bond-breaking and reformation, and charging (1, 2, 17). For example, the lifetimes of carriers can be relatively long and can produce secondary electrons while propagating through the specimen. These excitations can lead to irreversible bond breaking, especially if enough energy is present for significant atomic motion to occur (2). Delivering dose in a timed, highly-regular
15 manner may allow for a higher degree of carrier relaxation and bond reformation between arrival of each incident electron. For phonon excitation and specimen heating, arrival of multiple electrons in a short amount of time may result in accumulation of thermal energy, ultimately leading to irreversible structural disorder (*e.g.*, bond breaking, increased chain mobility, and melting). Thus, as with electronic transitions, damage of this type may be reduced by controlling
20 the timing of each electron such that complete relaxation and heat dissipation occurs between individual electron-excitation events.

Bond breaking and radical formation is also less likely to produce irreversible structural damage if the species lack sufficient energy to diffuse away from the initial reaction zone and

also if atomic vibration is minimized. By precisely timing the arrival of each electron, heat will not accumulate to a significant degree from repeated phonon excitation and frequent exothermic bond reformation. Further, diffusion rates will not become elevated, and thermal energy will be such that fewer species overcome activation barriers. Thus, the structure is more likely to be preserved due to recombination events frequently occurring at the site of initial bond breaking (17). In addition, observation here of increased damage for the conventional low-dose approach indicates that, despite being administered with the same dose rate, the random nature of the emission process can result in the arrival of multiple electrons at the specimen over relatively short periods of time. Following the arguments above, one would then expect these events to result in more frequent irreversible structural damage from otherwise reversible processes owing to increased accumulation of thermal energy and a higher spatiotemporal number density (*i.e.*, species per volume per time) of reactive species during moments of greater instantaneous dose.

One Electron Per Packet, 5 μ s Between Each Electron. To test the above-stated hypotheses, a set of pulsed-beam experiments were conducted, wherein both the time between electrons (f^{-1}) and the number of electrons in each packet were systematically varied and compared to the random thermionic-beam method and to one another. Figure 3 shows the result of reducing f^{-1} to 5 μ s ($f = 200$ kHz) for 1.04 ± 0.11 electrons per packet, again compared to the random-beam approach administered at the same dose rate (in this case, $9 \times 10^{-6} \text{ e} \cdot \text{\AA}^{-2} \cdot \text{s}^{-1}$ owing to a higher f). A statistically-significant reduction in damage at a given accumulated dose (here, up to $0.077 \text{ e} \cdot \text{\AA}^{-2}$) is again observed for the pulsed beam. Interestingly, damage for the pulsed-beam approach is again reduced by nearly a factor of two relative to the conventional low-dose method. The damage at a common accumulated dose (*e.g.*, $0.066 \text{ e} \cdot \text{\AA}^{-2}$), however, is higher by approximately 15% for both the pulsed and the random beams when compared to that sustained

with the lower dose rate shown in Fig. 2. This supports the hypothesis that contributions by irreversible aspects of otherwise reversible mechanisms to the accumulated damage sensitively depend upon the time between arrival of each individual electron at the specimen.



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Fig. 3. Comparison of damage induced by pulsed and random (thermionic) electron-beam irradiation for the same dose rate ($9 \times 10^{-6} \text{ e}\cdot\text{\AA}^{-2}\cdot\text{s}^{-1}$). Fading curves and least-squares fits for the pulsed and random beams are shown. The slope of the line fit to the random-beam data is steeper than that of the pulsed beam by a factor of 1.9. Pertinent specifications are as follows:

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Pulsed-beam current and illuminated area were $31.7 \pm 3.54 \text{ fA}$ and $219.4 \text{ }\mu\text{m}^2$, respectively.

Random-beam current and illuminated area were $32.0 \pm 3.45 \text{ fA}$ and $220.7 \text{ }\mu\text{m}^2$, respectively.

The pulsed beam was generated with a 300-fs FWHM pulsed laser operated at $f = 200 \text{ kHz}$ ($f^{-1} = 5 \text{ }\mu\text{s}$). The laser pulse energy was set such that the number of electrons per packet was measured to be 1.04 ± 0.11 .

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Increasing the Number of Electrons Per Packet. The effect of the number of electrons in each packet on damage was also studied, the results of which are summarized in Fig. 4. Because two factors were tested (f^1 and the number of electrons per packet), a two-level factorial approach was taken in order to avoid conflation. It was observed that the degree of damage sustained at a given accumulated dose is significantly higher when the number of electrons per packet is increased (here, from one to five; Fig. 4A). Reducing f^1 for the 5-electron packets from 100 μs to 5 μs produced a further increase in damage, potentially to a larger degree than for the 1-electron packets. An analysis of variance shows that both factors are statistically-significant predictors of damage (to 95% confidence), but no super-additive effect exists. That is, the results do not indicate that varying both factors produces any additional effect beyond the additive ones observed. This said, an unexplored region of the very-large parameter space may indeed exist where super-additive effects do occur. Interestingly, the deleterious effects caused by increasing the number of electrons per packet are substantially more significant than those caused by decreasing f^1 ; the five-fold increase in electrons per packet produces a greater increase in damage than a five-fold decrease in f^1 .

The dramatic increase in damage caused by a modest increase in the number of electrons per packet suggests that, for a given f^1 , a point should be reached where all benefits provided by pulsing the source compared to the conventional low-dose approach are lost. Figure 4B shows a comparison of pulsed- and random-beam irradiation, where the number of electrons per packet for the pulsed beam was increased to 20 ($f^1 = 5 \mu\text{s}$; same dose rate = $1.8 \times 10^{-4} \text{ e} \cdot \text{\AA}^{-2} \cdot \text{s}^{-1}$). As can be seen, no statistical difference (to 95% confidence) at a given dose (here, $0.063 \text{ e} \cdot \text{\AA}^{-2}$) is present, indicating that the amount of damage caused by the arrival of 20 electrons at the specimen over an estimated 2-ps time period [increased from 300 fs due to Coulomb repulsion

(44)] is comparable to that occurring during the conventional low-dose approach. It is reasonable to speculate that the quantitative behaviors observed here are material dependent, but more work is needed. Taken together, these results support the hypothesis that, despite being administered at the same dose rate, the statistical generation of multiple electrons over a short period of time during conventional emission processes is the source of the observed increased damage compared to the precision pulsed-source approach.

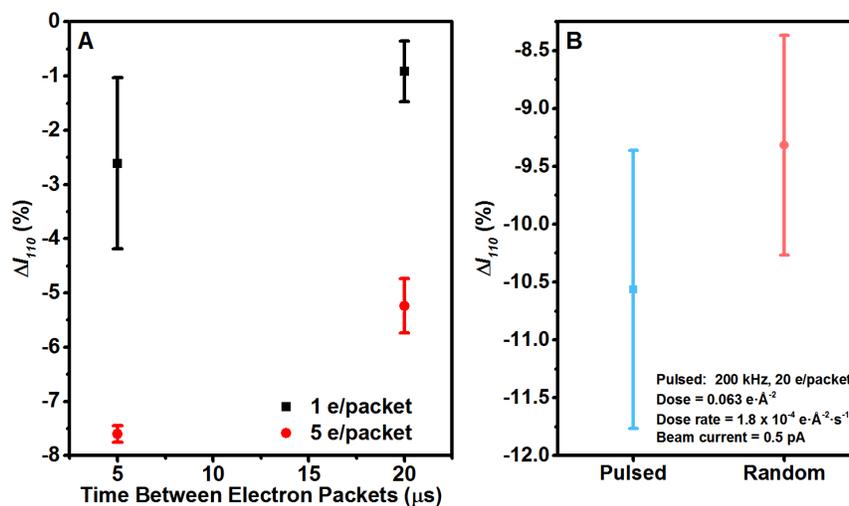


Fig. 4. Effect of the number of electrons per packet and the duration between each packet on damage. (A) Change in I_{110} after a total accumulated dose of $0.063 \text{ e}\cdot\text{\AA}^{-2}$. Error bars represent the standard error over four separate experiments. All data points were generated using a pulsed beam, and the same size specimen area was irradiated for each (within 0.5%). (B) Change in I_{110} after a total accumulated dose of $0.063 \text{ e}\cdot\text{\AA}^{-2}$ for a pulsed beam with 20 electrons per packet and $f^1 = 5 \mu\text{s}$ ($f = 200 \text{ kHz}$) compared to a random (thermionic) beam of the same current (0.5 pA) and for the same size irradiated specimen area (within 1%; same dose rate = $1.8 \times 10^{-4} \text{ e}\cdot\text{\AA}^{-2}\cdot\text{s}^{-1}$). Error bars represent the standard error over five separate experiments.

Summary and Conclusions

Via systematic studies and direct, statistical comparisons to conventional methods, we have conclusively shown that delivering dose using a precise, highly-controlled pulsed-beam approach in a TEM can reduce the damage sustained by beam-sensitive materials. We accomplish this by using the highly regular and stable emission from a fs pulsed laser to strictly control critical aspects of electron emission from the source. We find that the degree of damage is sensitive to both the duration between arrival of each electron at the specimen and the number of electrons per packet. Increased damage with increased electrons per packet, as well as the observation of a loss of advantageous effects, suggests the temporally random emission occurring during conventional low-dose methods is a significant contributor to beam-induced damage and that the pulsed-beam approach provides for an enhanced recovery process. The results of this work open an entirely new avenue of research into fundamental studies of radiation-damage mechanisms and into mitigation of the associated deleterious effects. Myriad future studies are envisioned, including quantifiably testing the effects on spatial resolution and extension of the approach to probing biological structures, biomaterials, biological macromolecules, liquid environments, and cryo-EM.

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materials availability: All data is available in the main text or the supplementary materials.

Data is available upon reasonable request from the corresponding author.

Supporting Information:

Materials and Methods

Figures S1-S8

Table S1

Supporting Information for

Reducing Radiation Damage in Soft Matter with Femtosecond Timed Single-Electron Packets

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This file includes:

Materials and Methods

Figs. S1 to S8

Table S1

Materials and Methods

TEM and Laser

All experiments (thermionic and pulsed photoelectron) were conducted with a 200 kV FEI Tecnai FEMTO transmission electron microscope (Thermo Fisher) equipped with a Gatan OneView CMOS camera. The Tecnai FEMTO is equipped with a conventional thermionic electron gun and Wehnelt triode. A custom graphite-encircled LaB₆ cathode (Applied Physics Technologies) was used for all experiments. The LaB₆ is truncated such that the emissive surface is flat and 50 μm in diameter. The graphite ensures that emission occurs mainly from the flat LaB₆ region and not from the shank, and it seems to also improve photoemission stability compared to bare LaB₆ cathodes, though experiments still need to be conducted to explicitly test this. The Tecnai FEMTO is equipped with an optical periscope integrated into the side of the column near the electron gun region (Fig. S1). This periscope provides optical access to the LaB₆ cathode such that femtosecond (fs) laser pulses can be used to generate discrete packets of photoelectrons from a room-temperature (*i.e.*, not heated) source. The fs laser used here was a Light Conversion PHAROS (6 W average power; Yb:KGW solid-state laser with regenerative amplifier; 1.03- μm fundamental wavelength, 1.2-eV photon energy). Photoelectrons were generated with fourth-harmonic light (4.8-eV photon energy, UV) generated by doubling 515-nm light produced using a Light Conversion HIRO harmonics generator. Laser pulse duration (300 fs full-width at half-maximum, FWHM) of the 1.03- μm fundamental was measured using a Light Conversion GECO scanning autocorrelator. This duration represents a lower bound of the UV pulse duration due to dispersion introduced during harmonic conversion and delivery to the TEM.

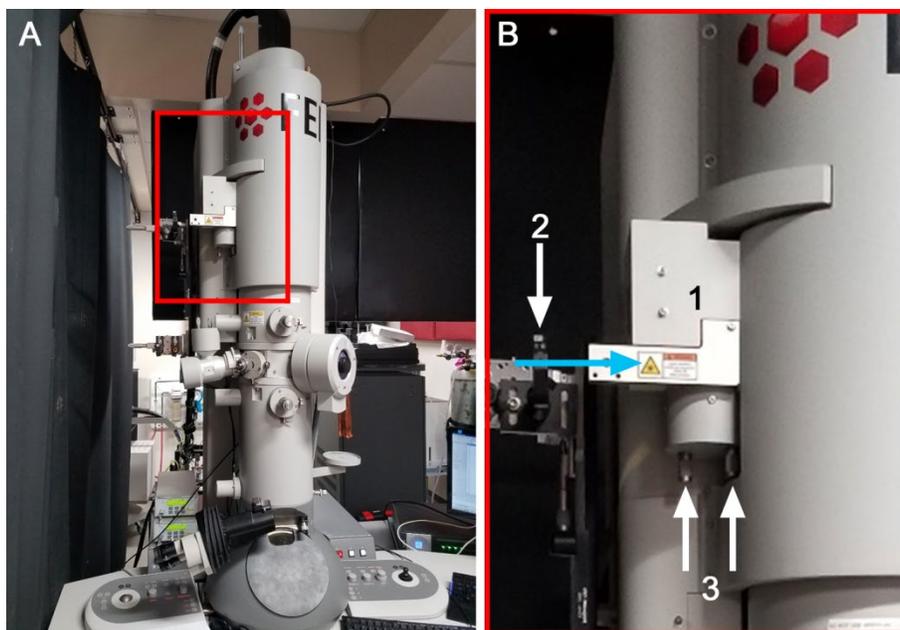


Fig. S1. Thermionic 200 kV TEM modified for laser-driven pulsed operation. (A) Photograph of the modified TEM highlighting (red rectangle) the optical periscope with which the fs UV laser pulses are directed onto the LaB₆ electron source. The fs laser system is behind the laser safety curtain to the left outside the frame of the image. (B) Close-up view of the optical

periscope highlighted in (A). Denoted in the image are: (1) The optical periscope assembly. (2) An external focusing lens mounted on a 3-axis positioner. (3) Micrometers for manipulating one of the internal mirrors. Direction of the incident UV pulses is indicated by the light-blue arrow.

Controlling Beam Current

Every effort was made to ensure imaging conditions were consistent across all experiments, and especially for directly-compared experiments. Thermionic beam current was controlled primarily using the cathode heat-to value. At times, spot size (*i.e.*, the C1 condenser lens setting) was changed slightly to fine-tune the beam current, but this was kept as close to the largest spot size of one as possible in order to ensure consistency and to maintain similarity to the relatively large photoelectron beam spot size used here. Pulsed photoelectron beam current was controlled by varying the fs UV laser pulse energy, which was used to finely control the number of electrons generated per laser pulse (Fig. S2), and *via* the laser repetition rate (f , pulses per second), which was used to control the number of discrete electron packets generated per unit time. While the overall linear behavior shown in Fig. S2 is typical when operating well below the space-charge (*i.e.*, saturation) limit (1-3), the precise values of the slope and the intercept can be highly variable from one LaB₆ cathode to the next, and even from one experiment to the next. Though the factors are likely numerous and highly synergistic, it is reasonable to assume the sources of variability include the changing condition of the cathode surface (*e.g.*, composition, morphology), variation in the laser focusing conditions on the cathode, variation in laser alignment onto the cathode, *etc.* Additional details specific to Fig. S2 are given in the caption. In light of this, care was taken to ensure each individual experiment, and each set of directly-compared experiments, were self-consistent. For example, the photoelectrons per laser pulse were measured and adjusted both before and after every experimental run, and the experiment was rejected and repeated if the difference was larger than the random error of the measurement. No reliance was placed on a predetermined quantitative relationship (*i.e.*, a universally applied calibration curve) between the UV laser pulse energy and the measured number of photoelectrons generated per pulse. Details regarding measuring beam currents are provided in the Measuring Beam Current section.

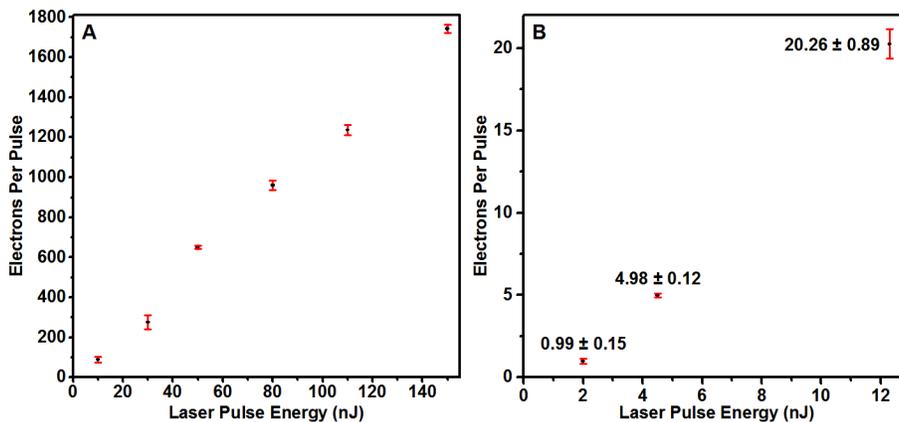


Fig. S2. Photoelectrons per laser pulse generated from the TEM LaB₆ source as a function of pulse energy. (A) Representative behavior of the photoelectrons per pulse for high pulse energies relative to those used to conduct the ultralow-dose-rate experiments. This plot is provided only as a general example of the overall behavior. (B) Photoelectrons per pulse at the

low pulse energies used for the ultralow-dose-rate experiments reported here. It is important to note that while the data shown in each panel are self-consistent, variations across the data sets are present. For example, in (A), the number of electrons generated per pulse for a laser pulse energy of 10 nJ was 89 ± 14 , while in (B), the number was 20 ± 1 for a pulse energy of 12.3 nJ. The reason for this is that the data sets were acquired in separate trials. Though the qualitative behaviors will be consistent (*e.g.*, linear response in the relatively low pulse-energy regime), variations in LaB₆ photocathode condition, precise laser alignment onto the photocathode, and the precise laser focusing condition onto the photocathode all are expected to cause variation in the absolute photoelectron values. For all experiments, the incident photon energy was 4.8 eV (fourth harmonic of the 1.03- μm fundamental wavelength of the Yb:KGW fs laser), and the pulse duration was 300 fs FWHM for the fundamental beam (*i.e.*, lower bound for the UV pulses). The error bars represent one standard error over ten separate measurements.

Experimental Parameters

Table S1 is a collection of pertinent parameters associated with all experiments presented and discussed in the main text. The experiment number corresponds to the figures in the main text; for example, experiment 2.1 is the first experiment associated with Fig. 2, and experiment 4A.1 is the first experiment associated with Fig. 4A. Note that in all cases, the beam area error is less than 1%. See the section Beam Area Determination for details of these measurements.

Exp.	Beam Type	TBP (μs)	E/P	E/P Error	Beam Current (fA)	Beam Current Error (fA)	Beam Area (μm^2)	Dose Rate ($\text{e} \cdot \text{\AA}^{-2} \cdot \text{s}^{-1}$)	Acq. Time (s)	Total Time (hrs)	Final ΔI_{110} (%)
2.1	Pulsed	100	1.03	0.15	1.6	0.24	132.4	7.76E-7	90	22.4	-0.81
2.2	Therm	n/a	n/a	n/a	1.6	0.22	131.5	7.77E-7	90	22.3	-1.55
3.1	Pulsed	5	1.04	0.11	33.3	3.45	218.9	9.52E-6	10	1.82	-0.93
3.2	Therm	n/a	n/a	n/a	32.2	3.71	218.6	9.20E-6	10	1.89	-1.66
4A.1	Pulsed	5	4.96	0.16	158.7	4.97	98.7	1.00E-4	5	0.173	-4.89
4A.2	Pulsed	5	5.02	0.05	160.8	1.64	98.8	1.02E-4	5	0.171	-5.60
4A.3	Pulsed	5	0.95	0.13	30.4	4.23	98.4	1.93E-5	5	0.898	-0.52
4A.4	Pulsed	5	0.89	0.14	28.5	4.56	98.7	1.81E-5	5	0.961	-1.31
4A.5	Pulsed	20	0.99	0.06	7.9	0.50	98.4	5.02E-6	5	3.46	-1.53
4A.6	Pulsed	20	1.08	0.10	8.6	0.78	98.9	5.47E-6	5	3.18	-3.75
4A.7	Pulsed	20	5.04	0.05	40.3	0.37	98.7	2.55E-5	5	0.680	-7.49
4A.8	Pulsed	20	4.96	0.06	39.7	0.52	98.6	2.52E-5	5	0.690	-7.70
4B.1	Pulsed	5	20.15	0.61	644.7	19.38	221.8	1.82E-4	2	0.096	-9.45
4B.2	Pulsed	5	19.71	0.56	630.6	18.07	221.1	1.78E-4	2	0.097	-9.72
4B.3	Pulsed	5	19.54	1.30	625.2	41.72	220.4	1.77E-4	2	0.098	-14.79
4B.4	Pulsed	5	20.32	1.13	650.2	36.17	220.1	1.85E-4	2	0.094	-11.25
4B.5	Pulsed	5	19.53	0.75	624.8	23.99	221.3	1.76E-4	2	0.098	-7.62
4B.6	Therm	n/a	n/a	n/a	627.5	16.92	220.8	1.78E-4	2	0.098	-9.86
4B.7	Therm	n/a	n/a	n/a	632.4	24.01	220.4	1.79E-4	2	0.097	-6.48
4B.8	Therm	n/a	n/a	n/a	636.9	15.99	220.9	1.80E-4	2	0.096	-7.82
4B.9	Therm	n/a	n/a	n/a	652.0	19.32	221.4	1.84E-4	2	0.094	-11.59
4B.10	Therm	n/a	n/a	n/a	649.1	18.60	220.0	1.84E-4	2	0.094	-10.85

Table S1. Experimental parameters and associated errors for all experiments presented in the main text. Exp. = Experiment number, where the first number is the figure number in the main text, and the second number is the specific experiment referred to. Beam Type = pulsed beam (Pulsed) or random thermionic beam (Therm). TBP = time between electron-packet arrival at the specimen. E/P = electrons per packet. Beam Area = size of the electron beam. Acq. Time = camera exposure used to acquire each diffraction pattern. Total Time = the total time required to conduct the fading-curve experiment up to a predetermined total dose. Final ΔI_{110} = the final average intensity value of the 110 Bragg spots up to the total accumulated dose specific to the experiment.

Photoelectron Beam-Current Stability

Instrument and lab-environment stability were especially critical to the successful execution of all experiments reported here, as the lowest-applied dose rates required over 22 continuous hours of data acquisition. Thus, long-term stability of the electron beam and the laboratory room temperature were regularly checked and continuously monitored. If the beam current (pulsed or thermionic) at the conclusion of a data-acquisition run was different from that at the start by more than the measured random error, or if the lab temperature changed by more than 1 °C during the experiment, the data set was rejected and the experiment was repeated. Lab temperature was continuously monitored at three locations: at the lab door, on the fs laser table, and at the TEM column. Changes of more than 1 °C at any of these locations at any point during the run resulted in rejection of the entire experiment. To illustrate the high levels of long-term instrument and lab stability that were reached here, Fig. S3 shows a representative plot of photoelectron counts (measured every 15 minutes by acquiring an image of the condensed beam) as a function of time over a 24-hour period. In this data, two standard deviations from the mean corresponds to a range of $\pm 0.3\%$ from the mean, and a drop in counts of 26 electrons per hour (or 0.0016% of the mean value) was detected, well below the range of standard deviation arising from random error.

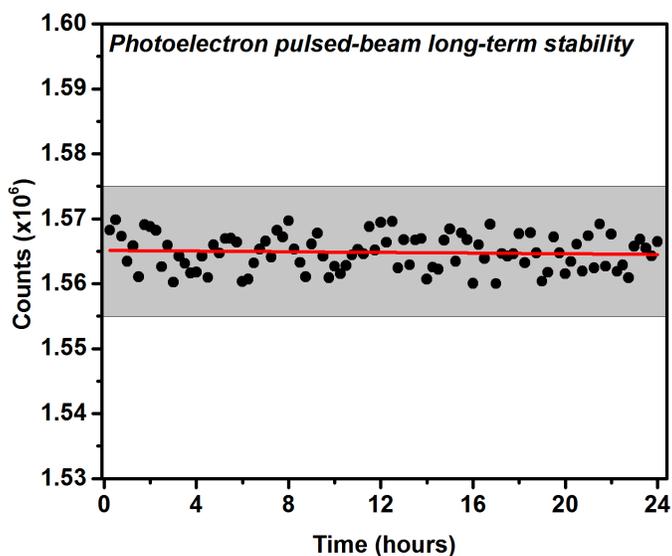


Fig. S3. Representative example of the long-term stability of the pulsed photoelectron beam over a 24-hour period. The data were obtained by summing the counts of an image of the electron source acquired at 15-minute intervals and then converting to the number of electrons incident on the detector. The mean value is $1.565 \pm 0.005 \times 10^6$ counts (electrons), where $\pm 0.005 \times 10^6$ (*i.e.*, $\pm 0.3\%$) is two standard deviations from the mean. The slope of the least-squares fit line (red) is -25.7, signifying a drop of 25.7 counts per hour, or approximately 0.0016% of the mean over 24 hours. To further emphasize the high long-term stability, the grey box denotes a $\pm 0.64\%$ deviation from the mean.

Bragg-Spot Fading Curve Method

A decrease of 110 Bragg-spot intensity as a function of dose and dose rate was used to track and quantify beam damage. An example of the type of data generated using this method is shown in Fig. S4. In this case, a hexatriacontane ($C_{36}H_{74}$, C36) microcrystal was irradiated with a 0.57 nA thermionic (random) beam, and a series of diffraction patterns (DPs) were acquired at steadily increasing dose. Progression of the damage manifests as a loss of intensity in the Bragg spots, with the higher-order spots being more sensitive than those generated at lower scattering angles (*i.e.*, larger planar spacings). The average intensity of spots generated from a family of planes is then plotted as a function of dose in order to quantify a damage rate. The dose, D , is $D = (I \cdot t) \cdot A^{-1}$, where I is the beam current ($e \cdot s^{-1}$), t is the exposure time (s), and A is beam area (\AA^2). For each experiment described in this work, a single C36 crystal was centered in the field of view, and DPs were acquired at regular intervals up to a predetermined total dose (*e.g.*, $0.063 e \cdot \text{\AA}^{-2}$). The predetermined dose was selected by balancing the need for high stability with the need for a sufficient amount of reliable data for making comparisons and drawing conclusions.

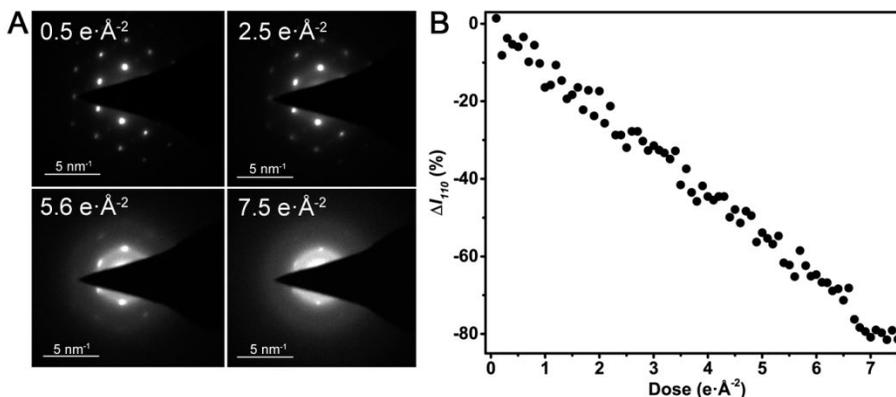


Fig. S4. Bragg-spot fading-curve method for monitoring beam-induced damage as a function of accumulated dose. (A) Representative diffraction patterns (DPs) from a C36 microcrystal as a function of dose ($e \cdot \text{\AA}^{-2}$). (B) Difference in intensity of the 110 Bragg spots as a function of dose relative to the intensities at nominally zero dose. Here, $\Delta I_{110} = (I_{110} - I_{0,110})/I_{0,110}$, where I_{110} is the average 110 Bragg-spot intensity at a non-zero dose, and $I_{0,110}$ is the average intensity at nominally zero dose.

Once generated, fading curves were least-squares fit with a line, and the resulting fit equation was used to calculate the value of ΔI_{110} at the predetermined dose (*e.g.*, $0.063 e \cdot \text{\AA}^{-2}$), as shown in Fig. S5. Here, the fading curves typically displayed a linear behavior, but in some cases the observed decay was exponential. Such non-linear behavior could be due to the

presence of residual solvent in the crystal (*i.e.*, incomplete solvent evaporation prior to conducting the experiment). Hydrocarbon contamination arising from degradation or polymerization of the small organic solvent molecules could potentially form a protective coating around the crystal or lead to an overall increase in thickness, which could affect the rate of damage in the specimen over time (4, 5). It may also be that certain damage, relaxation, or dynamic morphological mechanisms (*e.g.*, strain relaxation) dominate the overall behavior, depending upon the precise structural and compositional nature of the crystal or the manner in which it is interacting with the substrate. More work is needed to determine the source of the observed, though infrequent, deviation in linear behavior. Interestingly, the value of ΔI_{110} at the predetermined dose does not seem to be affected by the overall functional form of the fading curve. This was tested by completing a paired *t*-test on experimental data, with one set displaying linear-decay behavior and the other displaying exponential-decay behavior. The *p*-value on this paired *t*-test was 0.5, indicating no significant difference between the data generated from a linear *versus* an exponential fading-curve behavior at 95% confidence.

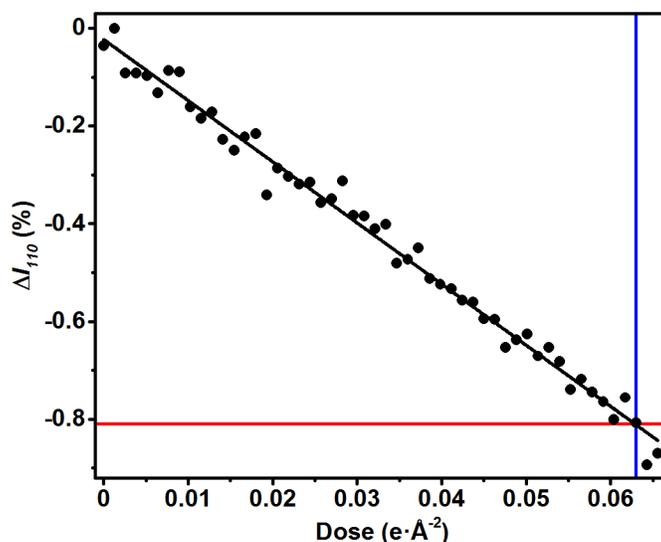


Fig. S5. Method for determining the value of ΔI_{110} at a predetermined total dose (here, 0.063 $e \cdot \text{\AA}^{-2}$). The vertical blue line marks the position of a dose of 0.063 $e \cdot \text{\AA}^{-2}$, the black line is the linear least-squares fit to the data (black circles), and the horizontal red line marks the position of the value of ΔI_{110} at the predetermined dose, as per solution to the equation for the linear least-squares fit to the data.

Specimen Preparation

Hexatriacontane ($C_{36}H_{74}$, C36) microcrystals were used as a model material for several reasons. First, damage due to electron-beam irradiation has been previously studied, and a range of critical doses (*i.e.*, dose producing a Bragg-spot intensity drop of e^{-1} or approximately 37%) have been reported (6-8). This provides benchmarks and points of reference for results of both the thermionic- and the pulsed-beam experiments reported here. Second, microcrystals oriented along the [001] crystallographic zone axis and with easily-quantifiable dimensions suitable for study with TEM are readily formed *via* drop casting from an organic solvent (*e.g.*, decane) onto conductive amorphous carbon grids. In addition, the crystals so formed have a low intrinsic defect density and a nearly monodisperse thickness distribution (see below). Thus, the influence

of thickness on measured radiation damage is effectively normalized out for all experiments (9). These properties are ideal for identifying multiple crystals suitable for beam-damage studies in order to generate statistics and for precisely determining geometric dimensions. It should be noted that much care was exercised in accounting for elastic deformation, strain relaxation, and n -beam dynamical scattering in the crystals, as these effects will impact the measured Bragg-spot intensities (6). Specifically here, specimen preparation consisted of drop casting a solution of C36 (Fluka Analytical, 98% purity) in decane (Acros, 99+% purity) onto a copper grid with a 12-nm-thick amorphous carbon substrate (Electron Microscopy Sciences, Q225-CR4) followed by solvent evaporation in air. A conductive support grid was used in order to reduce the effects of beam-induced specimen charging (4, 10). This method produces well-dispersed C36 single microcrystals with a nearly monodisperse thickness of four unit cells, as measured with atomic force microscopy (AFM) and a Bragg-spot intensity-ratio method (described in [Thickness of the C36 Specimens](#)).

Measuring Beam Current

Two methods were used to measure beam current. Method 1 consisted of using a Faraday cup integrated into the specimen holder (Gatan 636.MA double-tilt liquid-nitrogen holder) and connected to a 486 Keithley PicoAmmeter. Method 2 consisted of using the Gatan OneView CMOS camera to determine the counts per unit time and then conversion of this value to a number of electrons per unit time *via* a known scintillator calibration factor; note the calibration factor for the OneView camera is integrated into the software such that the number of electrons in the image is displayed. While it is reasonable to assume that the beam current measured with Method 1 can be directly related to the beam current used to irradiate the specimen, the geometry of Method 2 is such that the relationship between the beam current at the specimen and the beam current at the detector must be determined. This was accomplished by determining the ratio of the number of electrons per unit time (N) measured with Method 2 (detector; $N_{\text{det.}}$) and Method 1 (Faraday cup; N_{Faraday}) at several laser pulse energies (and therefore several beam currents). The results of such an experiment for $N_{\text{det.}}/N_{\text{Faraday}}$ are shown in Fig. S6.

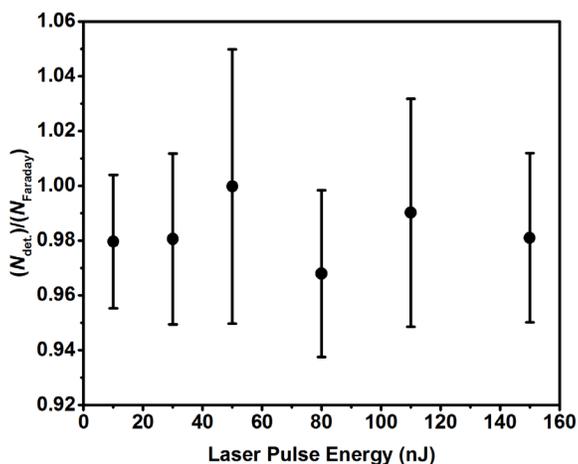


Fig. S6. Number of electrons per unit time measured at the detector ($N_{\text{det.}}$, Method 2) relative to the number of electrons measured at the specimen with a Faraday cup for the same amount of

time (N_{Faraday} , Method 1) for a range of laser pulse energies incident on the TEM photocathode. Error bars represent the standard error over 10 separate measurements.

Figure S6 demonstrates that, for the same lens settings and projection-system conditions, approximately 98% of the electrons measured at the specimen plane with the Faraday cup are incident on the detector. A t -test at all laser pulse energies tested indicates there is no statistical difference between the two measurement methods to 95% confidence. Note that the t -statistic between the ratio data in Fig. S6 and the value of 1 does not correspond to $p < 0.05$, indicating to 95% confidence that there is no statistical difference between the two forms of measurement. Also note that the overall variability was dominated by random error in the Faraday cup measurements (Method 1); measurements with the Faraday cup had up to 5% standard error, while those using the detector (Method 2) had a standard error of less than 1%. The result in Fig. S6 and the statistical equivalency to 95% confidence indicates it is reasonable to approximate the beam current measured at the detector as the beam current incident on the specimen. Another critical benefit to using Method 2 is the significantly increased sensitivity relative to the Faraday cup and picoammeter; the low random error and high sensitivity relative to the Faraday cup method enabled accurate and precise measurement of the number of electrons per laser pulse – down to the lowest of one electron per packet studied here.

Method 2 was used to monitor beam-current stability and to determine dose and dose rate for all experiments reported here. This was specifically accomplished by condensing the beam such that it fell entirely on the detector followed by acquisition of ten images with the same exposure time as the experiment of interest. The total counts for each image was obtained using Digital Micrograph 3. Ultra-Value Analysis was used to identify outliers in counts caused by (for example) X-ray detection. This resulted in rejection of no more than one image in a particular set of 10 (typically all 10 could be used). For each of the usable images, the total electron counts were divided by the image acquisition time to get the electrons per unit time, from which the mean and standard error were determined. The number of electrons per each individual packet could then be determined by noting the laser repetition rate (f), which dictates the number of discrete packets generated per unit time. See Table S1 for a summary of values and associated errors of pertinent experimental parameters.

Beam Area Determination

Because the electron-beam size may influence the observed damage behaviors (11-13), and because it is a key parameter for calculating dose and dose rate, it must be determined accurately and applied uniformly for direct comparisons to be made. For some experiments, the use of a small beam size (*via* a small condenser aperture) enabled the beam area to be directly measured. This was done by acquiring an image of the beam at the relevant intensity value and using a Hough transform to calculate the radius. When the beam shape was not circular, an edge-finding algorithm and numerical integration was used. When area could not be determined using a Hough transform or numerical integration (*e.g.*, because portions of the beam fell outside the detector), three additional methods were used (Fig. S7). For one method, an image of the beam was acquired in M magnification mode at all intensity values where the entire beam was visible on the detector, and the area was determined as described above. The relationship between intensity and beam size was then plotted and fit with a polynomial. Another method consisted of repeating this process but in LM magnification mode, enabling a wider range of intensity values to be directly measured. The difference in beam size between the LM and the M

modes was determined, and the average value was used to scale the LM sizes to the M mode. A third method consisted of comparing total counts for a beam imaged entirely on the detector to the counts for a spread beam. By comparing the counts in these two measurements, the proportion of the spread beam falling on the screen can be determined. The area of the spread beam can then be calculated using geometrical arguments despite not being entirely captured on the detector.

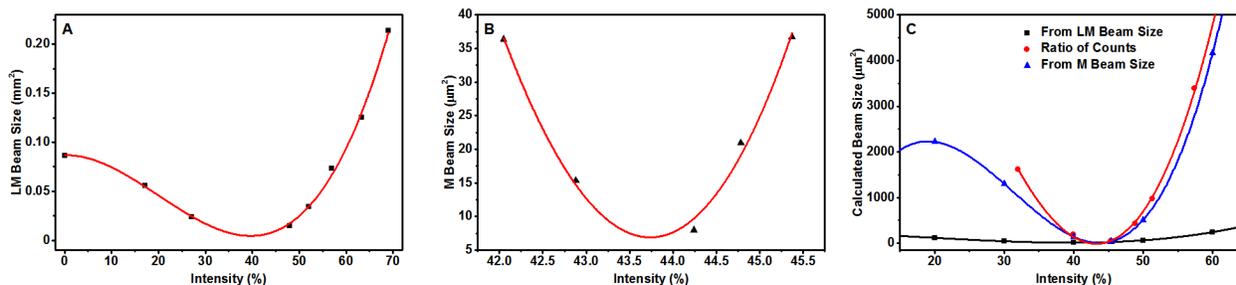


Fig. S7. Methods for determining the electron beam size. (A) Directly measured beam size for a range of intensity values in LM magnification mode (black squares). A polynomial fit to the data is shown in red. (B) Directly measured beam size for a range of intensity values in M magnification mode (black triangles). A polynomial fit to the data is shown in red. (C) Calculated beam size as a function of intensity value for three different methods over the intensity range of interest: (1) Fit to directly-measured LM-mode data (black). (2) Fit to directly-measured M-mode data (blue). (3) Geometrical approach based on the ratio of total to measured counts (red).

As can be seen from Fig. S7(C), there is good agreement between the M magnification method and the ratio of counts method. The LM magnification method was therefore not used. By comparison with similar measurements done with smaller condenser apertures (and therefore under conditions wherein a larger range of intensity values could be directly measured), it was consistently found that both of the retained methods were accurate to within the error of the counts measurement and the Hough transform – about 2% and 1%, respectively – at the intensity values used in all experiments (*i.e.*, 50% to 60%). Thus, the intensity ratio method for beam-size determination was used for all experiments owing to the ease with which multiple measurements could be quickly performed. This method was typically repeated several times during a given experiment and at least once at the beginning of every run. It is important to note that treating the beam shape as circular was found to be a reasonable assumption when a condenser aperture of 200 µm or smaller was used, which was the case for all experiments reported here.

Thickness of the C36 Specimens

Specimen thickness may affect the damage rates in some organic crystals (9, 14, 15). Thus, determining the thicknesses of the C36 microcrystals used here was deemed critical for making direct comparisons between pulsed- and thermionic-beam experiments. However, owing to the extreme beam sensitivity and softness of the microcrystals, it is generally challenging to directly measure thickness either during or immediately before the beam-damage experiment without modifying the structure in a way that could affect the outcome of the measurement. Measuring crystal thickness after the beam-damage experiment also poses a challenge due to potential mass loss, though this is likely to be minor owing to the low doses and low dose rates

studied here. A useful and important feature of C36 microcrystals is that thickness varies by integer multiples of the c -axis unit-cell length (6, 16, 17). Thus, intact crystals will vary in thickness by $n \cdot (c\text{-axis length})$, where $n = 1, 2, 3, \text{etc.}$

In order to rule out any influence of thickness on observed differences in damage between the pulsed and thermionic beams, a variety of methods were used to determine the distribution of thicknesses of C36 microcrystals formed *via* the preparation methods described in Specimen Preparation. Thus, the overall likelihood of conducting experiments on crystals of significantly different thickness was established. One method involved measuring the thicknesses of a large number of C36 microcrystals using atomic force microscopy (AFM). The crystals were prepared in the same manner as those used in the beam-damage studies (*i.e.*, same methods as described in Specimen Preparation and on the same type of TEM grid used in the beam-damage experiments). The center thicknesses of 107 individual, isolated (*i.e.*, non-overlapping) microcrystals were recorded. Of this set, 106 were found to be 19.1-nm thick, while one was found to be 23.8-nm thick, thus indicating the specimen preparation methods produce a nearly monodisperse thickness distribution.

Other methods employed here made use of the behavior of Bragg-spot intensities with thickness, mainly following work by Dorset (6). One such approach relies on the systematic change in Bragg-spot intensity ratios of the 110 and 200 families of planes with C36 crystal thickness (*i.e.*, number of stacked unit cells along the c -axis direction). Though mainly a qualitative measurement, it does allow one to determine if two specific crystals of interest are of approximately the same thickness. For example, for the pulsed and thermionic diffraction patterns shown in Figure 2 of the main text, the I_{200}/I_{110} ratios were 1.12 and 1.18, respectively, indicating the thicknesses were nearly the same. Dorset has also shown that an n -beam dynamical description is most appropriate for modeling Bragg-spot intensities from C36 crystals (6, 7). Indeed, the results of this work demonstrated that forbidden peaks readily appear in C36 crystals that are at least 24-nm thick. As no forbidden peaks were observed in the diffraction patterns analyzed in the beam-damage studies reported here, it is likely that all specimens were less than 24-nm thick, as indicated by the AFM measurements.

Dynamical scattering can contribute to allowed Bragg spots as well. Further, because the structure factors of particular reflections for C36 are more affected by n -beam scattering than others (*e.g.*, the 220 and 310 compared to the 120 , respectively), and because such scattering is a function of thickness, analyzing intensity ratios of such Bragg spots is another way to determine crystal thickness (6). The results of applying this method here are summarized in Fig. S8. By comparing calculations of Bragg-spot intensity ratios from data reported in (6) to the experimental data reported in Fig. 2 of the main text (for example), it was found that the thicknesses for both C36 crystals match, within error, the expected values arising from crystals that are 19-nm thick. Taken altogether, the results of the methods used here make it reasonable to conclude that the vast majority of C36 microcrystals formed using the described preparation methods were of uniform thickness (*i.e.*, 19 nm), and that the likelihood that beam-damage experiments were conducted on microcrystals of differing thicknesses is very low.

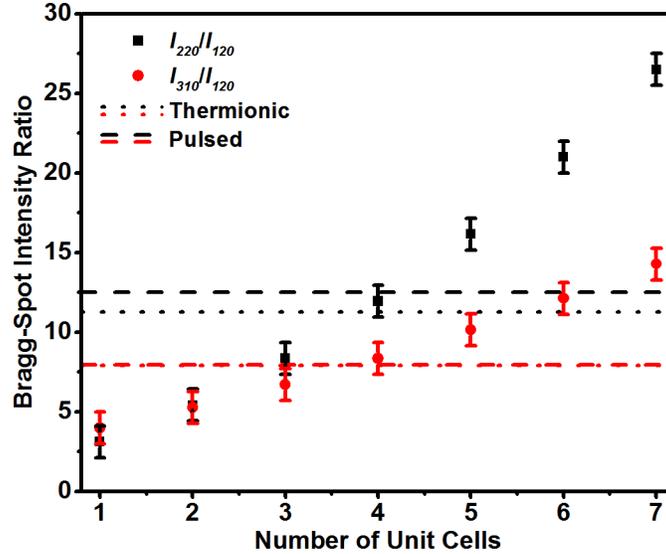


Fig. S8. Measured and calculated Bragg-spot intensity ratios for select reflections from crystalline C36. Individual data points represent the values of I_{220}/I_{120} (black squares) and I_{310}/I_{120} (red circles) as a function of C36 crystal thickness (in number of unit cells) calculated from data reported by Dorset (6). The error bars represent the estimated standard error based on the accuracy of the analysis technique and the estimated accuracy of digitizing the data from the source graph. The horizontal lines correspond to the intensity-ratio values measured here from the diffraction patterns shown in Fig. 2 of the main text, as generated with a thermionic (dotted) and a pulsed (dashed) beam. Note that red horizontal lines corresponding to the I_{310}/I_{120} ratio overlap with one another.

Data Analysis

After acquisition of all data and recording of all experimental parameters, the data analysis proceeded as follows. First, any shifts in position of the diffraction patterns was corrected using a template-matching plugin in Fiji (18) to calculate the shifts (in pixels in the x and y directions) and a MATLAB code to carry out the correction. This typically resulted in the loss of a few columns and rows of pixels around the frame edges, though no critical Bragg-spot information was lost. Next, the 110 Bragg peaks (the peaks of interest here) for all diffraction patterns in the stack were fit with a 2D Gaussian function, which included background determination. The intensities of the peaks were determined *via* background removal followed by numerical integration. Because the patterns were analyzed as a stack, the robustness of the automated fitting algorithm was routinely checked. This involved comparison of background values (which did not vary by more than 1% across all patterns) and comparison of peak fits for the first and last patterns, as well as those at all three quartiles, thus ensuring consistency of background correction and peak fitting across the entire stack. Finally, the intensities were plotted as a function of dose ($e\cdot\text{\AA}^{-2}$), and the resulting plots were fit either with a linear or a decaying exponential trend line. The total change in intensity for each peak was then calculated for a predetermined accumulated dose (*e.g.*, $0.063 e\cdot\text{\AA}^{-2}$). Note that when peak fitting with a 2D Gaussian was not ideal, which occasionally occurred for patterns generated with pulsed beams due to, for example, a non-circular laser profile on the cathode, an edge-finding algorithm was used to calculate the peak area.

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