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Optically Stimulated Luminescence (OSL) of Tooth Enamel and its Potential Use in Post-Radiation Exposure Triage

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Abstract

Optically stimulated luminescence (OSL) properties of dental enamel are discussed with a view to the development of an in-vivo dose assessment technique for medical triage following a radiological/nuclear accident or terrorist event. In the OSL technique, past radiation exposure is assessed by stimulating the sample with light of one wavelength and monitoring the luminescence at another wavelength under the assumption that the luminescence originates from the recombination of radiation-induced charges trapped at metastable defects in the enamel and that the intensity of the luminescence signal is in proportion to the absorbed radiation dose. Several primary findings emerged from this research: (a) sensitivities varied considerably between different teeth and also between fragments of the same tooth, (b) OSL signals were found to decay rapidly during the first 12 hours after irradiation and slower afterwards, (c) the fading rate of the luminescence signal varied between fragments, (d) blue light stimulation yields greater sensitivity than infra-red stimulation, while the OSL signal obtained with a high-intensity pulsed green-light laser was found to be not correlated with the radiation dose. Significant challenges remain to developing a practical in-vivo technique including the development of calibration procedures and lowering minimum detectable doses.

Keywords

Biodosimetry; optically stimulated luminescence; OSL; radiation; retrospective dosimetry; tooth enamel

Introduction

Numerous authorities, agencies, and expert groups have recognized the need for techniques that can be used in the aftermath of unplanned exposures to ionizing radiation to separate individuals into groups based on an estimation of the dose received by those individuals. In particular, it is desirable to identify those persons that received ionizing radiation doses of a magnitude that can benefit from medical treatment or short- and long-term surveillance. The reader is referred to, for example, the report of the Department of Homeland Security Joint Interagency Working Group (JIWG, 2005), Alexander et al. (2006), and Simon et al. (2006). These and other reports have highlighted the need for biodosimetric methods that can estimate radiation exposure to individuals in the range of 1-8 Gy with a throughput of one assay per 5 min.

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For a measurement technique to be suitable for field deployment, certain minimum requirements must be met. Possibly most important, a dosimetric material must be available in practically all persons exposed and it must be sensitive to radiation in terms of providing a signal that can be measured. The measured signal must be a function of the absorbed dose and should be able to be determined with a high-enough precision to minimize misclassification between ranges of exposure defined for specific triage actions. Tooth enamel appears to fulfill these requirements and has been used as detector for retrospective dose assessment using the electron paramagnetic resonance (EPR) technique (see for example, Desrosiers and Schauer, 2001; IAEA, 2002; Zhumadilov et al., 2007). To-date, however, the application of EPR to retrospective dosimetry has been limited to extracted teeth measured in the laboratory (i.e., in vitro measurements). Dosimetric properties of tooth enamel have also been investigated using optically stimulated luminescence (OSL) though the number of publications to-date is dwarfed by the number on EPR (see Godfrey-Smith and Pass, 1997; Yukihara et al., 2007; Godfrey-Smith, 2008; Haverland et al., these proceedings).

While many triage cut-off limits are in the 2 Gy to 4 Gy range, a desirable target for the minimum measurable dose (MMD) for an in-vivo procedure is 100 mGy. Development of a technique and the equipment to realize those MMD limits would allow for higher doses to be measured with ease and with good precision. Moreover, detection limits below 1 Gy would allow for long-term health research activities in addition to accomplishing the requirements necessary for triage and medical treatment.

X-band EPR signals of tooth enamel have been reported for absorbed doses as low as 100 mGy in laboratory procedures (e.g., Güttler and Wieser, 2008; Wieser et al., 2008). Development of the technique for in-vivo applications is also underway (Swartz et al., 2005, 2007; Williams et al., 2007), but currently measurable doses in vivo are at least one order of magnitude higher than in vitro.

In the OSL technique, radiation exposure is assessed by stimulating the sample with light of one wavelength and monitoring the intensity of the luminescence at another wavelength. This luminescence originates from the recombination of radiation-induced charges trapped at metastable defects in insulating crystals, which exist in the hydroxyapatite of tooth enamel. Since the intensity of the luminescence signal is a function of the absorbed energy or radiation dose (Bøtter-Jensen et al., 2003), the OSL technique allows for dose estimation using a suitable calibration factor. The OSL technique has distinct advantages over both EPR and thermoluminescence (TL) techniques. Compared to EPR, the primary advantages are that no complex spectra deconvolution and interpretation are required and the measurement equipment is much simpler and potentially more compact and deployable in difficult field conditions. Compared to TL, no heating of the sample is required.

General advantages include the fact that wavelength, intensity, and stimulation procedure (e.g. pulsed vs. continuous wave) can be adjusted which allows for optimizing OSL to the specific properties of tooth enamel. Finally, measurements are rapid and can be made remotely from the tooth using optical fibers. Use of optical fibers (Huston et al., 2001; Polf et al., 2002; Klein et al., 2005) potentially allows the technique to be used for in-vivo measurements without the need for extracting the tooth (Pass et al., 2003), an important requirement for an acceptable method and for real-time monitoring.

Godfrey-Smith and Pass (1997) were the first to propose OSL measurements of dental enamel. They were able to observe dose-dependent infrared- (IR-) and green-stimulated signals from deproteinated and undeproteinated (natural) human tooth enamel after a 120 Gy gamma-exposure. Yukihara et al. (2007) designed a higher sensitivity OSL system and used green and blue lasers for stimulation to show a MMD for powdered enamel of the order of 4 to 6 Gy.

Similar results were obtained by Godfrey-Smith (2008) stimulating with blue diodes with comparable stimulation power. The latter author also observed that the OSL signal was not stable over time and estimated that the signal would decrease to zero within two days following irradiation.

In order to develop a practical in-vivo measurement technique with high sample throughput (i.e., with short measurement times) based on OSL, better understanding is needed about the response of human tooth enamel and how it varies among individuals as well as between teeth of the same person.

In this paper, luminescence sensitivities resulting from different stimulation wavelengths and light sources are discussed. The sensitivity of multiple fragments from a single tooth as well as fragments from different teeth was investigated. The long-term signal stability and the sample-to-sample variability were examined.

Materials and Methods

Human molar teeth were obtained through cooperation between the National Cancer Institute and the National Institutes of Health. Teeth extracted for medical reasons were used in this study and were provided with no individual identifying information. Because of requirements of anonymity, no information was available regarding the medical histories of the donors. The apparent health of the teeth varied considerably from relatively healthy (based on a visual inspection) to those with clear signs of decay.

Sample Preparation

All tooth samples were disinfected in a 6% sodium hypochlorite solution for 24 h before use. Teeth crowns were separated from the roots, and the enamel layer was separated by cutting 1.5 - 2 mm slices from the sides and tops of the crowns with a laboratory diamond saw (IsoMet 1000; Buehler, Lake Bluff, IL, USA)[1]. The IsoCut Plus cutting fluid was replaced with iced distilled water to prevent possible chemical contamination of the samples and to prevent resetting of the luminescence by localized heating. Dentine was removed from the slices with a motorized hand drill (Dremel Variable Speed Multi Pro; Dremel, Racine, WI, USA), until no further dentine was apparent to the eye. A final cleaning of the samples with acetone for 5 minutes in a sonicator removed metal residues from drilling and slicing.

Some enamel fragments were reserved for measurements while others were crushed using an agate mortar and pestle and sieved into grain sizes of < 75 μm and 75-250 μm. Both grains, about 1 mg mass and spread in a thin layer, and fragments, were placed into carefully cleaned steel cups. No adhesive was used to affix the material to the cups. Once a batch of sample cups had been prepared, the samples were placed in the thoroughly cleaned OSL reader and not changed in order to minimize potential contamination due to handling.

Equipment

All measurements were carried out using a commercial OSL reader (Risø TL/OSL-DA-15 reader; Risø National Laboratory, Roskilde, Denmark) described by Bøtter-Jensen et al. (2000). The built-in $^{90}Sr/^{90}Y$ beta source gave a dose rate of ~100 mGy s^{-1}. Optical stimulation was carried out with blue light emitting diodes (LEDs) (wavelength of 470 ± 30 nm) delivering 31 mW cm^{-2} to the sample. Infra-red (IR) stimulation was from an IR LED array at a wavelength of 875 ± 80 nm with 100 mW cm^{-2} delivered to the sample. For green stimulation, we used a

[1]Certain commercial equipment, instruments, or materials are identified in this paper to foster understanding. Such identification does not imply recommendation or endorsement by the authors or their organizations, nor does it imply that the materials or equipment identified are necessarily the best available for the purpose.

modified light-guide attachment to couple a pulsed laser (532 nm, pulse width 15 ns, 4 kHz, 60 mW average power) into the system. A laser line filter ensured that only the desired laser line reached the sample and laser suppression filters were used in front of the photomultiplier to avoid overexposure.

The intensity of the emitted luminescence was measured by a light detection system consisting of a bialkali photomultiplier tube (Thorn EMI 9635QB; Electron Tubes, Rockaway, NJ, USA) and 290-370 nm band pass filters (Hoya U-340; THK Photo Products.Inc., Long Beach, CA, USA).

Results

Stimulation Schemes and Luminescence Sensitivity

Godfrey-Smith and Pass (1997) used IR diodes with 40 mW stimulation power and green stimulation using filtered light from a quartz envelope tungsten filament lamp. Godfrey-Smith (2008) stimulated with 50 mW blue diodes. Yukihara et al. (2007) used green and blue continuous-wave lasers with 23 and 27 mW cm^{-2}, respectively. All authors worked with powdered tooth enamel. To compare results with this previous body of work, the dose responses of two teeth were first measured with IR and blue stimulation. Powdered samples from both teeth, as well as from a fragment from tooth no. 2, were used to investigate the influence of grain size on the intensity of the luminescent signal. The OSL from empty sample cups was also measured using IR or blue diodes, both with and without radiation exposure. No signal was observed in any of these cases. Likewise, no blue or IR stimulated signal was observed from unirradiated teeth samples. These null measurements ensure that neither sample holders nor unirradiated teeth emit a luminescence signal that might make interpretation of the radiation-induced signal difficult.

Irradiated samples were stimulated for 200s with either IR or blue light. This stimulation time was found sufficient in all cases to bleach the signal to background levels. No difference in signal intensity and shape was observed for measurements carried out at room temperature and at 37°C (body temperature). The signal measured by the photomultiplier tube (PMT) consists of the luminescence emitted by the sample, plus a background due to electronic noise and stimulation light that is scattered by the sample and sample holder and which subsequently leaks through the filters. The OSL signal is defined as the PMT counts for 20s of stimulation minus the background. The background is measured by stimulating the same sample for 20 s, after it has been bleached for 180 s.

The blue-stimulated OSL (hereafter called OSL) signal (Fig. 1a) shows a rapid initial decay and 20 s after the onset of stimulation has almost reached background level, for all doses up to 500 Gy. Similar results were obtained with IR stimulation (“IRSL”). In general, the decay rate of OSL signals is a function of the incident optical power and the incident wavelength. Normally one expects that shorter wavelengths correspond to higher photoionization cross-sections for release of the trapped charge. Hence, the observation that IR-stimulated and the blue-stimulated OSL signals decay at similar rates is an interesting but unexplained phenomenon.

The dose responses obtained by blue stimulation of five sample cups with fragments or grains from two teeth are shown in Fig. 1b. The range of initial linear increase of the signal with dose is on the order of 100 Gy and saturation was not yet reached at the largest given dose of 500 Gy, in accordance with previous observations (Yukihara et al., 2007). The dose responses of the IR stimulated signals (not shown) vary from sample to sample, but are linear up to 10 Gy. The sensitivity, i.e. the signal intensity per dose unit, is listed in Table 1 for powder and fragment samples. For a saturating exponential-type response the sensitivity can only be

defined in the linear region. It varies between tooth samples, but is higher for small grains and lowest for the fragment, independent of the stimulation wavelength. A plausible explanation is the larger surface-to-mass ratio for small particles gives more surface area for the stimulation signal, but the possibility cannot be excluded that the crushing process changes the sensitivity of the material. A higher sensitivity will also result in a smaller MMD. Blue stimulation results in higher signals and is therefore favored over IR stimulation.

Yukihara et al. (2007) were able to obtain MMDs of four to six Gy for green-stimulated OSL using a CW green laser. Pulsed stimulation (often called POSL) has been shown to achieve a higher sensitivity than CW-OSL in certain materials (Bøtter-Jensen et al., 2003). For that reason, we attempted to measure a dose response using a pulsed green laser with high stimulation peak-power. Samples were stimulated for 1200s or until a stable signal level was reached for each measurement. High signals were observed from tooth fragments without prior exposure to ionizing radiation in a manner that subsequent sequences of irradiation and measurement showed no correlation with ionizing radiation dose (Fig. 2). Instead, the signal was observed to decrease with each subsequent measurement, independent of the given ionizing radiation dose. This suggests a slow bleach of a higher, pre-existing signal and also indicates that the “stable” level reached after 1200 s still contains some slow decaying luminescence. In an attempt to correct for this pre-existing signal, the luminescence signal was calculated by subtracting the background obtained from the previous measurement instead of subtracting the background obtained from the same measurement, but to no avail. These measurements indicate that such a technique would not be useful at doses below about 30 Gy.

Dose Response and Tooth-to-Tooth Variability

To predict an absorbed dose with acceptable precision from an OSL signal, calibration procedures are necessary. In a conventional laboratory measurement situation, each tooth enamel sample is irradiated with one or several known doses from a radioactive or X-ray source after the initial OSL measurement. This dose response, i.e. the signals obtained after laboratory irradiation, provides an individual calibration curve that can be used together with the initial OSL signal to estimate the absorbed radiation dose. Since an in-vivo procedure will not allow radiation exposure of the tooth, an alternative approach was considered for deriving a calibration curve. The approach was to attempt to derive a “universal” dose response curve based on an average dose response from many teeth as is also done in EPR enamel dosimetry.

Measurements were made on 17 fragments from 11 different teeth. The fragments were weighed and their dimensions measured, to calculate an approximate surface area. They were then irradiated with known ionizing radiation doses and the blue stimulated signals were measured. Signals that were larger than three times the standard deviation of the background are plotted in Fig. 3a. They show a wide distribution of luminescence sensitivities, even for fragments from the same tooth. No correlation was found between the sensitivity and the apparent condition (healthy or decayed) of the teeth.

For each individual fragment, the dose response was fitted linearly. The measured signals were compared with the corresponding dose responses and the resulting doses were calculated, 40 in total. The deviation between these estimated doses and the true administered doses are listed in Table 2. The majority of all estimated doses were within 10% of the administered dose, most within even 5%. Few showed a deviation of more than 20% and none more than 30%. This confirms that doses to tooth enamel can be estimated with satisfactory precision, particularly when an individual calibration is available.

In a second step the signals for each dose value were averaged and an average dose response was determined (Fig. 3a). All 40 normalized signals were compared with this average dose response to test its suitability for estimating absorbed dose to individual teeth without further

calibration. The resulting deviations of these calculated doses from the given doses are again listed in Table 2. Less than half of the values show a deviation smaller than 50%; a significant number deviate even more than 100% from the given doses, which can be mainly attributed to the large differences in luminescence sensitivity. The histogram of the signals without any normalization is plotted in Fig. 3b. The standard deviation was 76% of the average value. The distribution is skewed with the maximum shifted to smaller than average values. The fragments were of different size and shape, but all smaller than the illuminated area. For that reason, we tested to see whether normalization of the signals by the weight or area of the fragments would improve the calibration. This testing used 50 Gy exposures. This normalization procedure was found to further enhance the skewness (Fig. 3b) and the standard deviation was larger than 100%. The results indicate that large signals do not necessarily originate from large or heavy fragments, but are caused by individual luminescence characteristics, which are conventionally monitored by controlled radiation exposure. Normalization by a signal obtained after irradiation with a 10 Gy beta dose resulted in a distribution around the average value with 32% standard deviation, a much improved result.

Signal Stability

Godfrey-Smith (2008) observed that the OSL signal obtained after blue stimulation (and measured in the UV emission range) was not stable over the first 2.8 h after irradiation. In that work, the OSL signal was estimated to decrease to zero within two days. We subsequently investigated two questions to better understand the fading phenomenon: (1) Is the rate of this “fading” sample dependent?, and (2) Is a stable level reached after a long waiting period?

Nine fragments from seven different teeth were irradiated with 80 Gy and the signal was measured immediately after irradiation. The experiment was repeated with increasing delay periods between irradiation and measurement. Samples were stored and measured at room temperature. OSL signals normalized to the signal measured directly after irradiation are plotted in Fig. 4. Fading rates vary between fragments from the same tooth as well as between different teeth, but, for all fragments, fading is rapid during the first few hours and progresses more slowly afterwards. After one hour only 60-80% of the initial signals are left and these values decrease to 35-60% after 12h (Fig. 4a). The difference between the fragments increases over time. The fading includes signal components that are thermally unstable at room temperature. In a laboratory procedure these components can be removed by holding samples at elevated temperatures for specific periods of time after irradiation. When the OSL signal is subsequently measured, thermally unstable signals will have decayed. Fig. 4b shows that the OSL signal from tooth enamel does not stabilize even after 48 hours. A measurement carried out 40 days after irradiation indicated additional signal reduction. The signal level at that time generally ranged from 0% to 30% of the original signal, although one sample seemed to have stabilized at a 60% signal level. Further and longer fading tests would be required to prove the stability of this signal level. A variation was also observed between fragments from the same tooth (see open and filled circle and square symbols).

As the signals decrease, MMDs increase with the delay between radiation exposure and measurement. The relative spread between fragments increases, resulting in a greater uncertainty for the measured doses. Correction factors for fading are conventionally obtained by measuring the signal decay over days or weeks (e.g. Auclaire et al., 2003), a procedure not feasible for triage situations.

Discussion and Conclusions

Tooth enamel shows a measurable OSL signal following irradiation when stimulated by continuous-wave blue or IR light. These OSL signals increase with dose. The sensitivity depends on the physical form of the enamel (in vitro) and is higher for fine powder than for

fragments. Possible explanations of the latter observation include the larger surface-to-weight ratio for the powder or perhaps other unknown sensitization processes related to crushing the enamel. Sensitivities depend on the stimulation light and the sample form, and are higher with blue stimulation than with IR stimulation. Based on the present technology used and the background PMT counts, current MMD estimates range from 1-5 Gy for blue stimulation to 10-15 Gy for IR stimulated signals. Improvements in technology may very well lead to improvements, lowering, of the MMD. The high peak stimulation power of a green pulsed laser resulted in a high signal from an unirradiated tooth and subsequent irradiations and measurements showed no correlation between signal and dose. Therefore, our experiments indicate that continuous wave stimulation with wavelengths in the blue region is the most promising of the investigated stimulation schemes.

Dose response curves varied considerably between different teeth and also between fragments from the same tooth. Attempts to use an average dose response as a universal calibration curve resulted in dose errors of more than 100% for some samples. Calibration by administering a known laboratory dose reduced the variation considerably, but is not suitable for in-vivo measurements. Hence, the results emphasize the need for a calibration procedure and to minimize the complexity of the in-vivo instrumentation, an all-optical calibration procedure would be preferable. Rudko et al. (2007) showed that UV and gamma radiation induce similar EPR signals in tooth enamel. The influence of UV radiation on the OSL signal needs to be investigated. If a UV-induced OSL signal proves to be proportional to the beta and/or gamma radiation sensitivity, samples can be calibrated by illuminating the tooth with a UV lamp and measuring the resulting OSL signal.

OSL signals were found to decay rapidly during the first 12 hours after irradiation and slower afterwards. The rate of this fading varied between the investigated fragments and fading continued after 2 days. However, while fading may introduce some uncertainty in the doses measured during a triage situation it may also prove to be advantageous if it can be demonstrated that the signal from previous medical exposures has faded and that the signal measured is only from the most recent exposure event. To do so it is clear that further investigations of signal fading, how it varies among teeth, and the influence of radiation type and energy are required.

The results of the presented experiments are a necessary step towards understanding the OSL characteristics of human tooth enamel and towards developing a stable and repeatable in-vivo measurement methodology. Our present success with measuring doses in the range of 1-5 Gy suggests that further research and improvements in technology may result in a field-deployable tool (as proposed by Pass et al., 2003) capable of rapid exposure assessment without many of the problematical requirements imposed by other methods.

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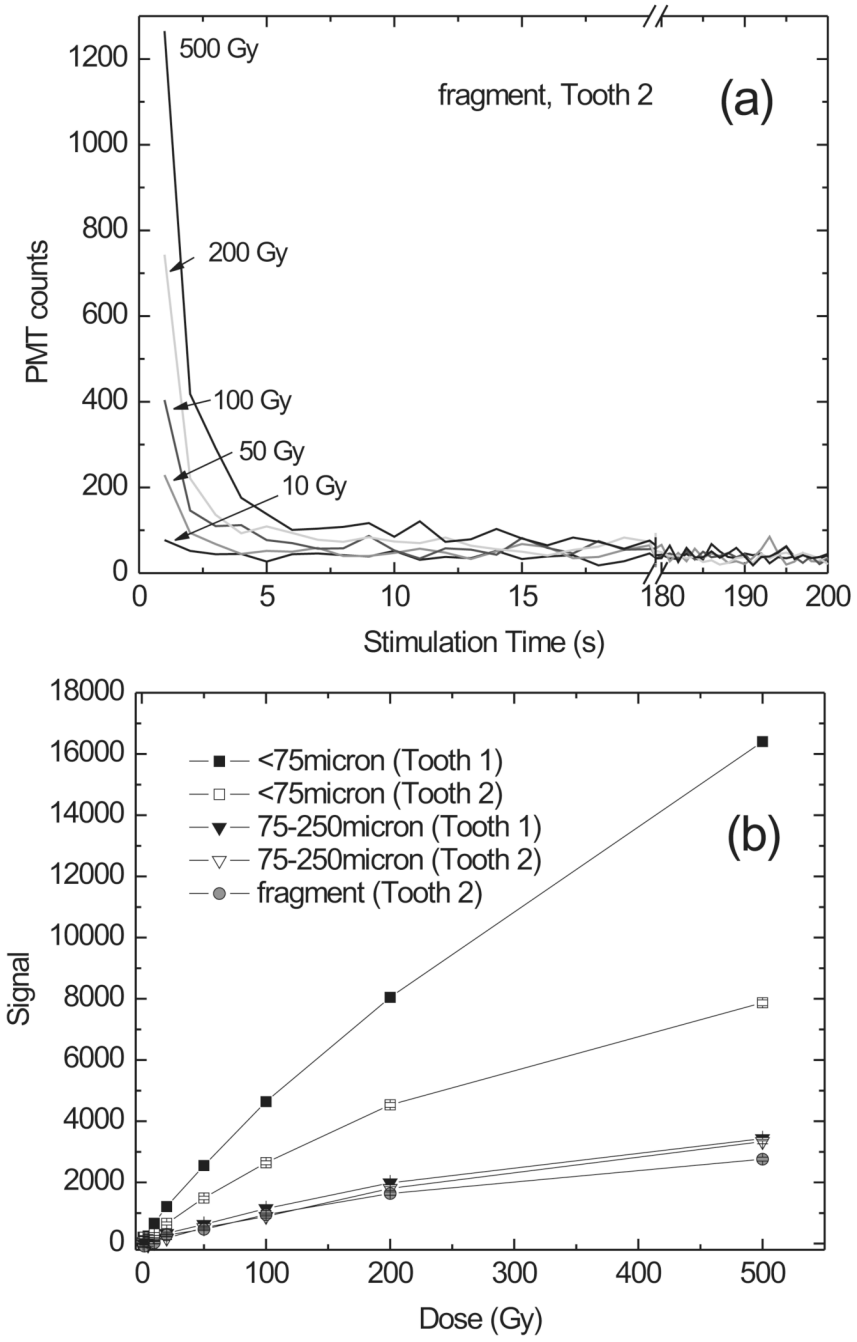

Fig. 1.

(a) Blue stimulated signal and background after irradiation with the doses listed in the figure legend for a fragment from tooth 2. (b) Dose response for grains and fragments from tooth 1 and tooth 2.

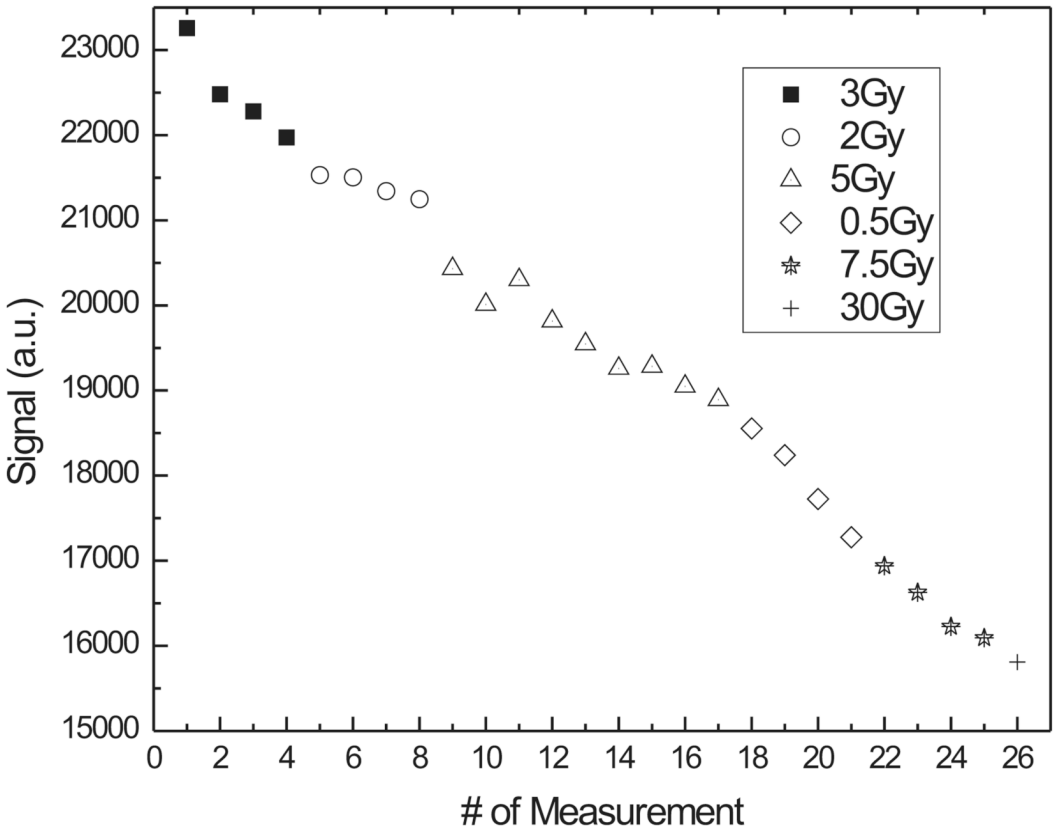

Fig. 2.
Signal obtained by stimulating with a green pulsed laser after the doses listed in the legend were given. The doses were delivered and the subsequent OSL measured in the sequence shown.

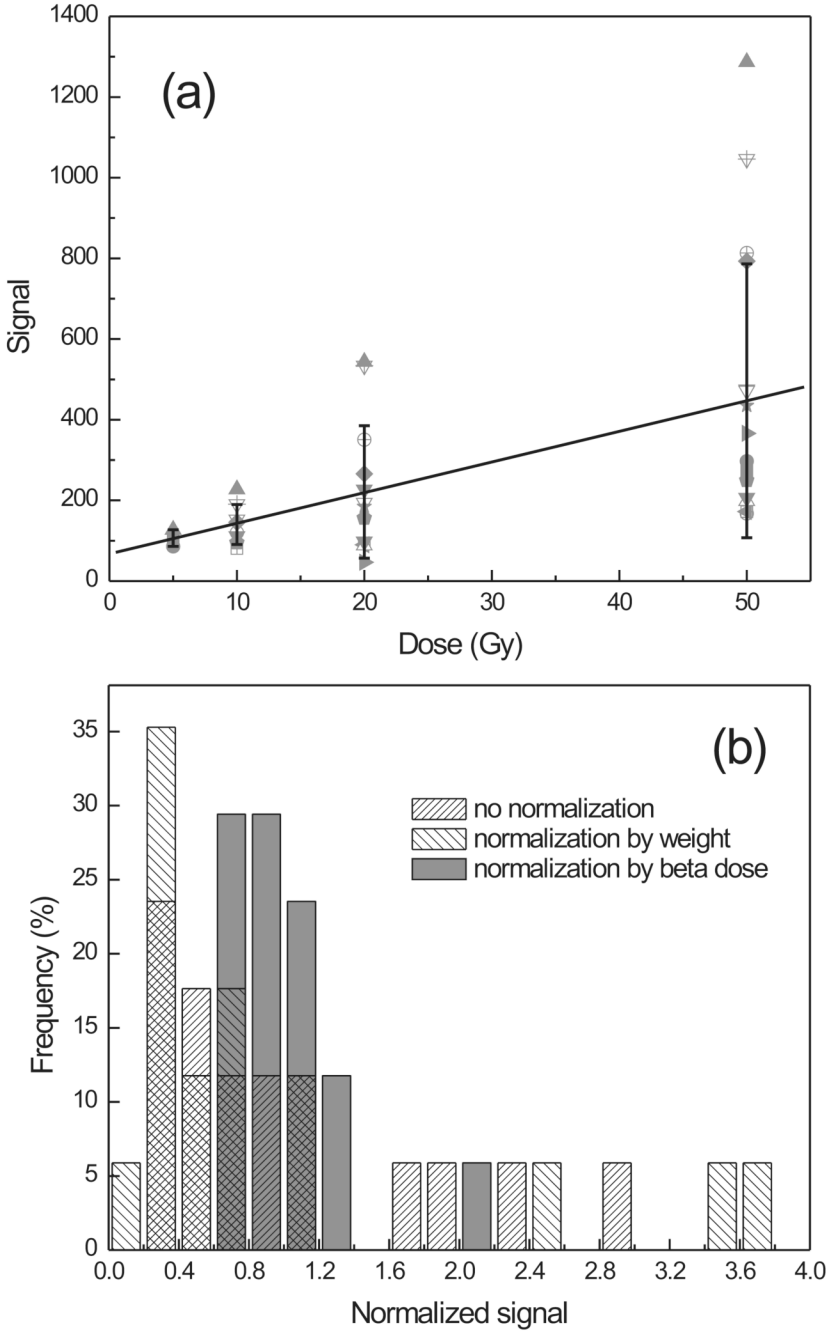

Fig. 3.

(a) Measured dose responses (symbols) for 17 tooth fragments from 11 different teeth. Similar symbols denote fragments from the same tooth; e.g. all downward pointing triangles denote one tooth, different fragments are distinguished by filled and open symbols. The average dose response is indicated by the solid line. The error bars reflect the standard deviation of the average signals. (b) Histogram for signals obtained by blue stimulation after irradiation with 50 Gy: (i) as measured, (ii) normalized by the weight of the fragment, (iii) normalized by the signal resulting from a 10 Gy dose. All distributions have been normalized to their mean values.

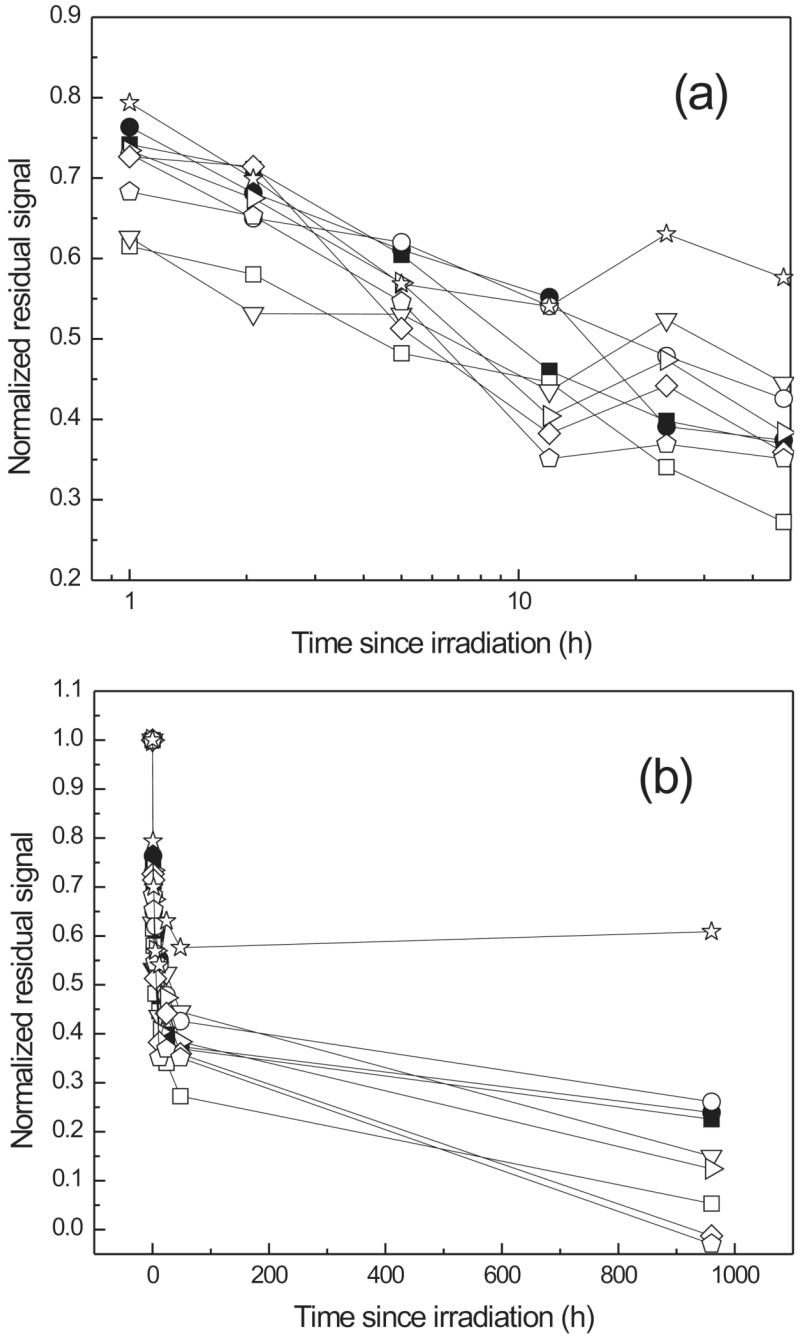

Fig. 4.

OSL signals of 9 tooth fragments, normalized to the signal measured directly after irradiation. Similar symbols denote fragments from the same tooth; e.g. all downward pointing triangles denote one tooth, different fragments are distinguished by filled and open symbols. (a) Short term fading with signals measured 1 h, 2 h, 5 h, 12 h, 24 h, and 48 h after irradiation. Note the logarithmic time scale (b) Long-term fading over 40 days.

Table 1

Sensitivity in counts Gy^{-1} for IR (IRSL) and blue stimulation (OSL) immediately following irradiation. Note the larger mass of the fragment compared to the powdered samples.

Tooth sample, grain diameter (μm), mass	IRSL sensitivity (counts Gy^{-1})	OSL sensitivity (counts Gy^{-1})
No. 1, < 75μm, 1mg	11	46
No. 2, < 75μm, 1mg	8	26
No. 1, 75-250 μm, 1mg	8	11
No. 2, 75-250 μm, 1mg	5	9
No. 2, fragment, >1mg	2	10

Table 2

Number of teeth deviating from administered dose stratified by percent difference between estimated dose and administered dose. Total number of teeth was 40. Measured signals were compared with the individual dose response for each tooth and an average dose response determined from the average signals for each dose value.

Dose response used	<10%	10-20%	20-50%	50-100%	>100%
Individual	29	8	3	0	0
Average	6	2	8	19	6