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INTRODUCTION:

The overall hypothesis driving this work was that an increase in microdamage activity during repeated loading of bone will signal the approaching fatigue fracture. The formation and propagation of microdamage, in the form of microcracks and diffuse damage, produces detectable acoustic emission events similar to the seismic motions of an earthquake. Therefore, impending fatigue fracture of bone can be detected by acoustic emissions. In order to test this hypothesis, fresh-frozen human tibias from males and females between the ages of 20 and 50 years old were acquired for use in testing procedure outlined below. Sections of the tibias were removed and machined into a proper geometry for fatigue tests. Microdamage formation with characteristic waveform properties was monitored through the use of piezoelectric acoustic emission transducers. The primary aim of this work was to investigate and quantify the damage events during the *in vitro* fatigue loading of cortical bone fatigue fracture model.

BODY:

The key objectives of this work were to: 1) determine the characteristics of damage events that signal the onset of fatigue fracture in bone and 2) quantify the differences between genders on the evolution of fatigue damage. Attainment of these aims required:

- a) Preparation of specimens from male and female tibias
- b) Mechanical induction of fatigue damage within these specimens through fatigue tests
- c) Monitoring microcracks using acoustic emission technique in real-time during fatigue loading
- d) Analysis of the data obtained from above tests
- e) Identification of any acoustic emission parameter(s) that may have the potential to detect the onset of a failure

Procurement of Tibias and Specimen Preparation

Since the focus of this work is the fatigue/fracture properties of those likely to be subjected to the rigors of military training, we needed to restrain our donor pool to a relevant age range. Beams were machined from tibia samples from fifteen different donors ranging in age from 22 years old to 49 years old, both male and female. These specimens were divided into four testing groups based on the criteria of age (young – old) and gender (male – female). There were four donors from each of these testing groups except for one group which had three. The younger group was taken from samples of up to the third decade. The older group was taken from donors of the fourth and fifth decade. These two age groups were then divided into male and female. There were 32 samples taken from the donors, with 8 samples coming in each of the four testing groups. When the beams were machined, the anatomical site of the beam in the tibia was noted and recorded. Each of the testing groups had comparable number of samples from each anatomical location. These different testing groups were designed so that it could be possible to observe trends in the data that related to age, gender, or location in the tibia.

Mechanical Induction of Fatigue Damage

We chose to use a three-point bending test configuration in order to induce fatigue damage to the specimens due to this configuration's ability to subject the specimens to both tensile, compressive and shear loading modes, which best reflects the richness of *in vivo* loads experienced by bones. Fatigue testing was carried out under load-controlled conditions and the decrease in specimen compliance was used as a measure of fatigue damage accumulation.

Monitoring Microcracks Using Acoustic Emission

The acoustic emission system consisted of two acoustic emission transducers, two signal amplifiers and a computer which analyzes and stores the AE data (Figure 1). Commercial preamplifiers, computer control board and software were used. New transducers that were small and robust enough for the planned test configuration were designed and fabricated in lab. Since irrigation of the bone specimens was an integral part of the experimental set up, these transducers were fabricated to withstand irrigation. The transducers were mounted on the specimen using cyanoacrylate glue. The acoustic emission signal from the transducers was preamplified (Physical Acoustics Corp., New Jersey) and acquired at a rate of 5 MHz using an acoustic emission system (Vallen Systeme, Munich, Germany). This system was able to record emissions when they exceeded a set threshold of 40.0 dB. Certain parameters of these emissions were recorded including amplitude, rise time, duration, counts and energy. The total activity of microdamage was assessed by recording the cumulative number of these acoustic emissions events. We were able to isolate any extraneous noise from the machine or motion of the fixtures and keep only the signals of interest by assessing the difference in arrival times of acoustic emission waves at these sensors. This was carried out using standard pencil lead (0.3 mm) lead breaks on the bone specimens.

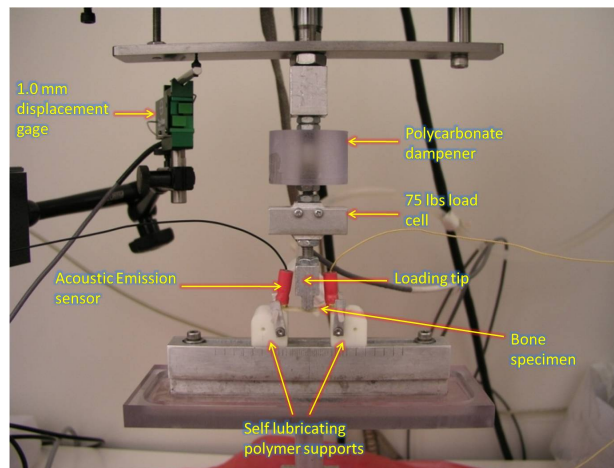


Figure 1: Three-point bending experimental set up

The tests which were conducted at the onset of our study used four-point bending configuration in order to check the test setup for determining the AE parameters during testing (Wasserman *et. al.*, Appendix I). Beam-shaped cortical bone specimens ($n = 4$) were machined from the diaphyses of femurs such that the longer axis of the beams were aligned along the longer axis of the mid-diaphyseal shaft. Fatigue testing was conducted at four different stress levels and the AE parameters were recorded until the specimen failed. The test machine recorded the load and displacement data from the load cell and machine actuator displacement, and a second compliance measure was determined by dividing the difference in maximum and minimum displacement by the difference in the maximum and minimum load (displacement/load). We observed that the increase in the rate of acoustic emission events was concomitant with the characteristic knee region of the compliance curve which shoots up early in the cascade of events which leads to failure. Figure 2 shows a typical curve. Results of this pilot study demonstrated that failure of standardized cortical bone specimens can be predicted ahead

of time by monitoring the rate of damage accumulation via the acoustic emission technique. The predictive ability of the technique was such that the failure was detected within $78\% \pm 15\%$ of the fatigue life on the average. However, we abandoned the four-point bending configuration because the bone specimens would break at the loading tips. Testing with the three-point bending setup showed the expected increase in compliance during fatigue loading in a consistent manner, thus justifying its substitute for four-point bending. Due to the length of time necessary to carry out the fatigue test (up to 14 hours), specimens were kept hydrated in a saline solution supplemented with CaCl_2 and protease inhibitors to prevent the degradation of the tissue.

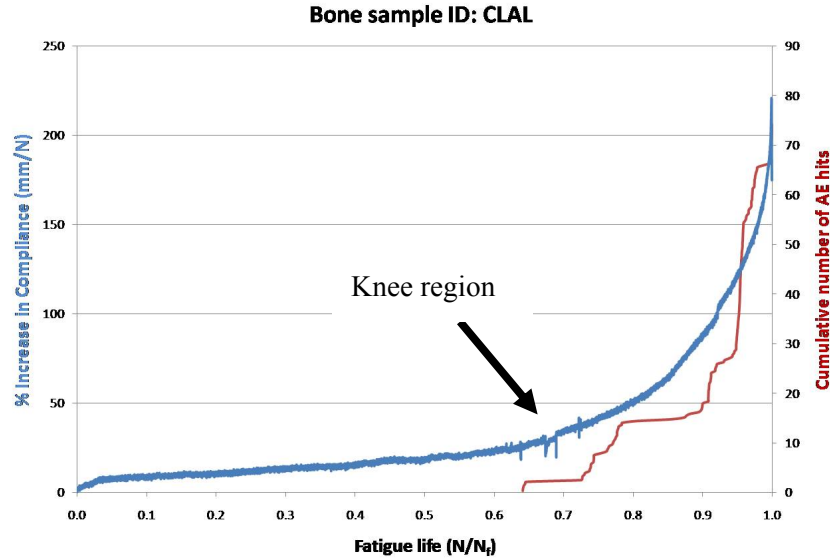


Figure 2: % Increase in compliance (mm/N) and cumulative number of AE hits vs. fatigue life; N: number of cycles and N_f : number of cycles at failure

While *in vitro* aspect of this study was under investigation, the preliminary results from the three-point bending test configuration were encouraging enough for us to start exploring the *in vivo* side which was beyond the scope mentioned in the grant proposal. We wanted to know whether or not in borne acoustic emissions from microcracks could make it to the surface of the skin. That, in turn, meant to evaluate the attenuation coefficients of muscle and bone. These were assessed by using broadband acoustic emission transducers in a pulse transmitter - receiver mode. Coefficients for both transverse and longitudinal directions were evaluated. In a similar fashion, *in vivo* attenuation coefficients were calculated to characterize the difference in attenuation between live and dead tissue (under Institutional Review Board (IRB) approval from Purdue University). Finally, since the ultimate vision is to prevent stress fractures in live subjects, the efficacy of a portable data acquisition system was assessed. Two experiments were performed on three healthy subjects (24 – 36 years old) (under IRB approval from Purdue University) with the portable data acquisition system turned on: 1) running a distance of 600 ft. and 2) hopping 60 times at one location.

KEY RESEARCH ACCOMPLISHMENTS:

- Acquisition of 16 donor tibias (some paired, some not) from which 32 samples were machined (for three-point bending test) using low speed diamond blade saw (Buehler, Illinois).

- Designed and built a three-point bending apparatus that:
 - Accommodates the specimen size needed for standardized testing
 - Allows the attachment of two miniature acoustic emission transducers
 - Allows specimen displacement monitoring from an external displacement gage
- Fabricated acoustic emission transducers and eliminated extraneous events which stem from activities other than the formation of microdamage.
- Conducted three-point bending tests on thirteen specimens, selected randomly.
- Analyzed the data collected for prefailure AE indicators; identified several indicators with varying degree of success.
- Feasibility of a portable acoustic emission system for *in vivo* use was also evaluated.
- Designed and characterized acoustic emission sensors specifically for *in vivo* application

Table 1 Percent fatigue life at threshold (FL) and time remaining for failure (TRF) for bone specimens using 53 dB AE amplitude as the threshold. Red rows indicate that the 53 dB criterion predicts the onset of failure too late, whereas the criterion worked well for other samples.

Sample	Sex	Age	FL(N/N _f)	TRF (mins.)
LLAL	F	38	0.825	154.7
BLMP	M	25	0.863	71.4
NLLP	M	22	0.932	55.7
ORAL	M	44	0.998	1.6
CLAL	F	31	0.908	15.3
FLAL	M	22	0.988	3.8
HLPL	F	25	0.985	6.5
CLMP	F	31	0.999	0.0
MRPL	F	49	0.887	11.0
ERPL	F	49	0.999	0.0
KRAM	F	24	0.996	0.5
NLMP	M	22	0.910	15.5
MRMP	M	49	0.786	106.3
Average		33.2	0.9	34.0
Stdev.		11.2	0.1	49.2

N: Number of fatigue cycles; N_f: Number of fatigue cycles at failure; FL (N/ N_f): Percent fatigue life spent by the detection of the AE threshold; TRF (mins.): Time remaining to failure

Table 1 was generated using the time at which the first emission exceeding 53 dB amplitude as the emission signaling the onset of failure. Figure 2 shows such a knee region for the compliance data from one of the bone samples. Using 53 dB amplitude of the acoustic emission as a sole characteristic feature, 8/13 specimens could have been prevented from the final failure using the 53 dB amplitude as the test-stop criterion. Red colored rows indicate the specimens whose remaining fatigue life is less than 5 minutes (i.e. failure predicted too late). Average time before failure by which the failure can be detected using this criterion is 34 mins. ($\pm$ 49.2).

Table 2 Time remaining for failure (TRF) under different AE parameters. Italicized values in blue indicate that a given threshold predicts the onset of an incipient failure *too early* in terms of its remaining fatigue life whereas the underlined values in red indicate that the same threshold predicts the failure *too late*.

AE parameter (Threshold)			Amplitude (53 dB)	Rise Time (8 μ -seconds)	Duration (50 μ -seconds)	Cum. AE hits* (150)	Rate of AE hits^ (2 sec ⁻¹)	Combination%
Sample	Sex	Age	TRF (min)	TRF (min)	TRF (min)	TRF (min)	TRF (min)	TRF (min)
LLAL	F	38	154.7	166.4	48.6	151.83	<i>861.0</i>	48.6
BLMP	M	25	71.4	202.5	170.8	80.6	91.8	71.4
NLLP	M	22	55.7	441.2	441.2	<i>756.3</i>	<i>758.6</i>	55.7
ORAL	M	44	<u>1.6</u>	<i>889.1</i>	<u>1.6</u>	<u>0.0</u>	<i>853.6</i>	<i>853.6</i>
CLAL	F	31	15.3	39.2	20.6	6.9	15.3	15.3
FLAL	M	22	<u>3.8</u>	7.7	7.7	<u>4.1</u>	<u>7.7</u>	7.7
HLPL	F	25	6.5	<i>416.2</i>	7.2	<i>419.6</i>	<i>424.6</i>	6.5
CLMP	F	31	<u>0.0</u>	<u>0.0</u>	<u>0.0</u>	<u>0.0</u>	<u>0.0</u>	<u>0.0</u>
MRPL	F	49	11.0	12.4	11.0	<u>2.1</u>	9.7	9.7
ERPL	F	49	<u>0.0</u>	<u>0.0</u>	<u>0.0</u>	<u>0.0</u>	<u>0.0</u>	<u>0.0</u>
KRAM	F	24	<u>0.5</u>	13.3	<u>0.5</u>	<u>0.0</u>	13.3	13.3
NLMP	M	22	15.5	<i>125.9</i>	15.5	<u>0.0</u>	15.5	15.5
MRMP	M	49	106.3	106.5	106.7	29.3	106.7	29.3

*Cumulative number of AE hits

^Rate of accumulation of AE hits

%Combination indicates the prediction of impending failure by invoking all AE variables and choosing the one which provided the prediction of incipient failure neither too early nor too late.

Table 2 summarizes the remaining fatigue life for specimens using all AE parameters i.e. rise time, duration, cumulative number of AE hits, rate of AE hits and a combination of all these parameters. In establishing this table, we assumed that a prediction was too late if it occurred within 5 minutes of the failure. Also, a prediction was accepted as too early if it happened earlier than 2 hours prior to the failure (120 mins.). When all the AE variables are invoked as such, the failure of 10 out of 13 samples can be predicted reliably (neither too soon nor too late). The three samples which could not be reliably predicted (*viz.* CLMP, ERPL and FLAL) from breaking happened to break within a very short span of time (less than half an hour).

REPORTABLE OUTCOMES:

- Performed tests to verify whether acoustic emissions signal the onset of failure prior to the final fracture and the results were presented at the 51st Annual Meeting of the Orthopaedic Research Society as a conference abstract (Appendix I).
- Energy-based characteristics of microfracture events during fatigue loading of human cortical bone specimens were evaluated and the results were submitted as a conference abstract to the Orthopaedic Research Society (Appendix II).
- Assessed the *in vitro* attenuation due to muscle and bone in the longitudinal and transverse directions and a conference abstract was submitted to the American Society of Mechanical Engineers – Summer Bioengineering Conference (Appendix III).
- Attenuation due to in-born acoustic emission events in a dissected dog limb and the source intensity of these events was assessed using microbeams. The results were presented at the 2007 BMES Conference (Los Angeles, CA) (Appendix IV)
- Characterized the *in vivo* attenuation due to musculoskeletal tissue and, thus, demonstrated the efficacy of the acoustic emission approach. Results were submitted to the Annual Meeting of the Orthopaedic Research Society (Appendix V).
- Demonstrated the feasibility of a portable acoustic emission system and lab made acoustic emission sensors in an *in-vivo* application and the results were submitted to the American Society of Mechanical Engineers – Summer Bioengineering Conference (Appendix VI).

CONCLUSIONS:

The goal of these experiments was to determine an indicator or a set of indicators from the acoustic emission technique which could predict the onset of failure before it occurs within a reasonable amount of time. The data acquired using cortical bone beams from distal regions of human tibia shows evidence of AE parameters such as rise time, duration, cumulative number and amplitude of acoustic emissions being the precursors of the final failure. Future work consists of doing these experiments on more specimens according to the protocol stated in the methodology section so as to populate the data for statistical significance. Although the number

of samples covered hasn't allowed for a meaningful comparison based on gender, these experiments are ongoing and will be completed and submitted as a manuscript by the end of summer 2008.

Having observed the data from the three-point bending test configuration, we took the initiative of experimenting with the *in vivo* aspect of this problem in parallel. Attenuation coefficients for muscle and bone *in vitro* were found out. Using amplitude and rise time indicators in combination allowed predicting impending failure reliably (i.e. neither prematurely nor too late) in nine out of thirteen samples. Further, such coefficients were calculated for the *in vivo* case as well. We also tested the efficacy of a portable acoustic emission system. It is believed that in its ultimate form, such a system would enable interception with the training regime prior to the incidence of the fracture as signaled by acoustic emissions. It would not only reduce the time necessary for recuperation but also increase the preparedness and effectiveness of military personnel.

Appendix I

ACOUSTIC EMISSION TECHNIQUE CAN PREDICT THE FATIGUE FAILURE

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INTRODUCTION

Stress fractures of bone constitute the most serious musculoskeletal overuse injury during military and athletic training (1). The cascade of events prior to the incidence of stress fractures involves the upregulation of bone turnover, amplification of porosity, induction of greater local strains and associated increase in damage activity. Therefore, we propose that it would be possible to diagnose the progression of the failure process by monitoring the microdamage activity. Acoustic emission technique detects the stress waves generated during the formation of microdamage using surface mounted piezoelectric transducers; thus, it is a viable alternative to monitor damage activity (2). More importantly, it is a non destructive and non invasive technique which has the potential to be utilized as an *in vivo* diagnostic tool. In the current study we sought to answer the following questions: 1) can the onset of stress fractures be predicted by monitoring the microdamage activity via acoustic emissions and 2) how soon before the actual failure can be predicted via acoustic emission. This hypothesis was investigated by listening to acoustic emissions released during *in vitro* fatigue loading of standardized specimens from human femoral cortical bone.

METHODS

Four prismatic beams were machined from the mid- femoral diaphyses of two males (52 and 53 years-old) using a low-speed saw (South Bay Tech). The widths and the thicknesses of beams were machined as allowed by the actual cross-sectional geometry of femurs, resulting in a thickness range of 2.5 mm to 3.3 mm and a width range of 6.5 mm to 7.5 mm. Lengths of beams were kept standard at 70 mm in length. Beams were subjected to fatigue in four point bending with an inner span length of 30 mm and outer span length of 60 mm under continuous irrigation of calcium supplemented saline solution (3). The cyclic loading was conducted under load control and the maximum load varied to create stresses corresponding to 60% to 80% of the yield stress which was obtained from prior monotonic tests of two prismatic beams. The minimum load was kept at $1/10^{\text{th}}$ of the maximum load. The loading waveform was triangular with a 0.1 sec long ramp down followed by a 0.1 sec long ramp up at the rate of 2 Hz. The manifestation of damaging events were assessed by calculating the compliance of specimens by dividing the strain range with the stress range as calculated from the load and displacement (as recorded at the loading point) values, respectively.

An acoustic emission transducer (Pico, PAC, NJ) was mounted at the mid-span of the specimens using cyanoacrylate glue. Signal from the transducer was preamplified and acquired at a rate of 2 MHz using a specialized acoustic emission system (AEDSP 32/16, PAC, NJ). The activity of microdamage was assessed by recording the cumulative number of acoustic emission events.

RESULTS

The specimens were loaded in the low-cycle fatigue regime and the number of cycles to failure (N_F) ranged from 600 to 35020 cycles (Table 1). Compliance curves exhibited two temporal stages: a region of stability where the compliance did not change notably and a second stage characterized by a knee region during which the compliance increased rapidly and concluded with failure of the specimen (Figure 1). Concomitant with the initiation of the knee region was an abrupt increase in the cumulative number of acoustic emission events indicating that prefailure events are predominantly highlighted by the microdamage activity.

We have taken the number of cycles at which the rate of accumulation of acoustic emissions begins (N_P) to spike as the cycle at which acoustic emissions predict the onset of failure. The capacity of acoustic emissions to predict failure was calculated in terms of the percentage of specimen's fatigue life, i.e. $(N_P/N_F)*100$. In all specimens precursor acoustic emission events were detected in the range prior to the characteristic knee region and these events were likely to stem from initiation of microdamage.

DISCUSSION

Results of the current study demonstrated that failure of standardized cortical bone specimens can be predicted ahead of time by monitoring the rate of damage accumulation via the acoustic emission technique. The predictive ability of the technique was such that the failure was detected within $78\% \pm 15\%$ of the fatigue life on the average. The current analysis has focused on the rate of accumulation of acoustic emissions only. Acoustic emissions are sinusoidal bursts and further valuable information could be extracted from these bursts such as the duration, amplitude, energy and the frequency content. These waveforms can be classified to identify and extract those bursts which mark the onset of failure (4). Further refinement of the method holds promise for *in vivo* detection of stress fractures in the field using acoustic emissions.

Table 1. The predictive capability of acoustic emissions expressed in terms of the specimen's fatigue life

	Maximum Stress [MPa]	N_F , Fatigue Life [cycles]	N_P , Fracture Onset via AE [cycles]	Predictive Capability [% of Fatigue Life]
Specimen 1	55	26363	22580	85%
Specimen 2	61	35020	33382	95%
Specimen 3	66	4737	2989	63%
Specimen 4	71	600	402	67%

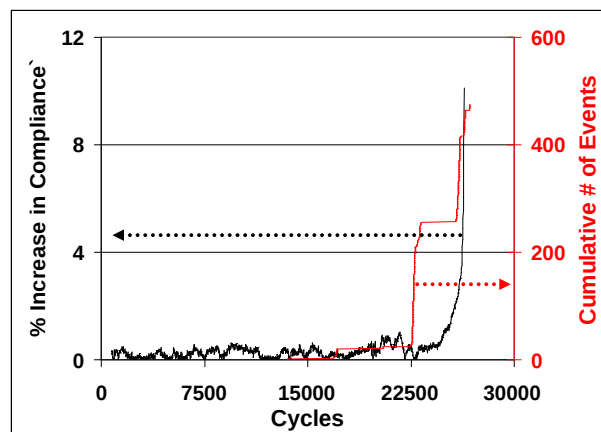


Figure 1. The variation of compliance and the cumulative number of acoustic emission hits with loading cycles.

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Appendix II

ENERGETICS OF MICROFAILURE DURING FATIGUE OF HUMAN CORTICAL BONE

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INTRODUCTION

Stress fracture of bone is a process which involves generation and propagation of microfailure events. Earlier studies have examined damage accumulation during fatigue testing at different intervals using histology (1). Histology provides substantial insight to damage evolution; however, it is an inherently static process and it can reflect the temporal changes in microfailure processes only by time-consuming histological processing. We have previously shown the ability of acoustic emission (AE, stress wave generated by microfailure events) to predict the impending fatigue failure by simply monitoring the rate of accumulation of acoustic emission events (2). In the current study, we further investigated the energy-based characteristics of microfailure events during fatigue failure of human cortical bone.

MATERIALS & METHODS

Three prismatic beams were machined from the distal diaphysis of tibias from two human donors (31 y.o. female, $n = 2$ and 1 x 25 y. o., $n = 1$ male) using a low-speed saw (South Bay Tech, CA). The widths and the thicknesses of beams were machined as allowed by the actual cross-sectional geometry of femurs, resulting in a thickness range of 1.1 mm to 2.7 mm and a width range of 2.8 mm to 7.2 mm. Lengths of beams were kept standard at 40 mm in length. Beams were subjected to fatigue in three-point bending with a span length of 25 mm and kept under continuous irrigation with calcium supplemented saline solution (3). An initial cyclic loading was conducted within the elastic range in order to determine the initial elastic modulus which was used to calculate the load necessary to create approximately 0.65% strain. Specimens were fatigued under sinusoidal loading with the minimum load corresponding to 10% of the maximum load at a frequency of 2 Hz until failure. The compliance of specimens was calculated from the load and displacement values measured at the mid-span. Compliance is a measure by which one can monitor the overall failure of the specimen and it increases asymptotically towards failure. Acoustic emission (AE) transducers were mounted to the outer supports and the signals associated with AE events were recorded using an AE data acquisition system (Physical Acoustics Corp., NJ). The energetics of microfailure processes were assessed by monitoring the duration, amplitude and energy of recorded AE signals.

RESULTS

The three specimens failed at 9800, 11,800 and 28,000 cycles and the increase in compliance at failure was 25%, 71%, and 126%, respectively. The larger the number of cycles to failure, the larger the number of acoustic emissions were detected (184, 656, 4612, respectively). During the course of fatigue process, acoustic emissions occurred in several bouts each of which were separated by periods of very low AE activity (Fig. 1a). These bouts of AE activity were concomitant with increases in compliance (Fig 1b). Prior to the increase in compliance a small number of low energy, low amplitude, and short duration acoustic events occurred (see circled highlight in Fig. 1b). Following the increase in compliance a large number of high energy, high amplitude and long duration acoustic events took place. The final failure was associated with events whose energy exceeded 35,000 dB- μ s, amplitudes exceeding 60 dB, and durations larger than 1000 μ s. Once events of these magnitudes were attained, the failure appeared to be imminent.

DISCUSSION

The current study demonstrated three consistent characteristics for accumulation of AE events: 1) damage accumulation occurred in isolated bouts during fatigue, 2) the energy contents of emissions varied throughout the test, and 3) very high energy events took place before the final failure. It is likely that each bout results from induction and growth of microcracks. This growth apparently resulted either in arrest or the growth increment was below the detectable level; thus, leading to periods of relative silence. The low energy, low amplitude, and short duration events prior to increases in compliance (Fig 1b) could arise from incomplete prefailure processes. As the microfailure takes place completely, the compliance increases and higher energy events

take place. These observations are in general agreement with previous histological analyses of fatigue damage in bovine bone (1). The future work will use histological analysis of these samples after the occurrence of AE bouts. This way we will be able to associate the energetics of AE events with the physical evidence of damage. This combined approach will shed light on the strength and weaknesses of bone tissue during fatigue and increase our understanding on the material level basis of stress fractures.

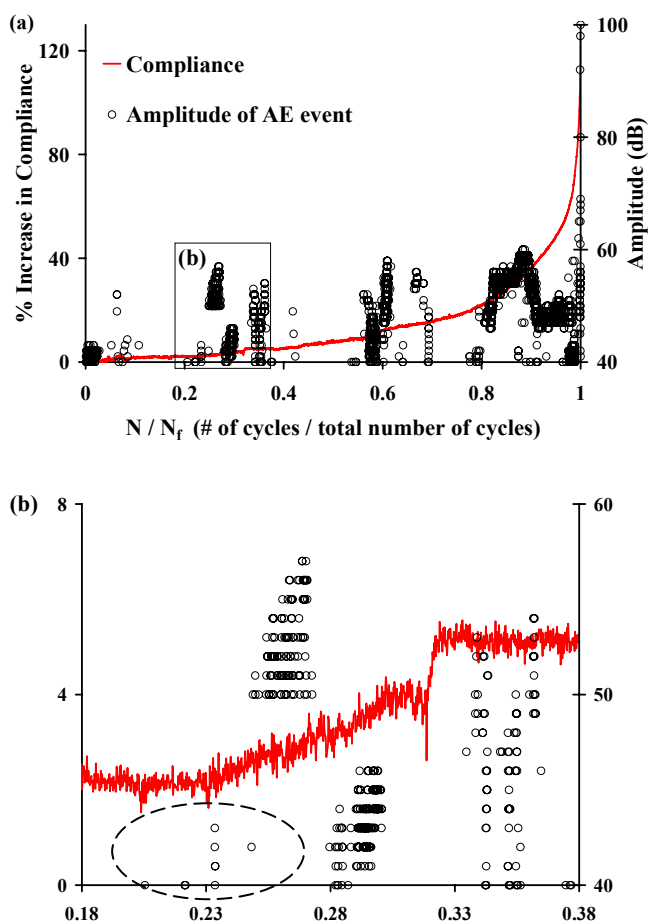


Figure 1: a) Plot of the increase in compliance (left ordinate) and the amplitude of acoustic events (right ordinate) in relation to the normalized number of cycles. Each circle represents one acoustic emission event. b) A close-up view of the inset in 'Fig 1a' where the compliance displaced a local increase.

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- (3) Gustafson MB, et al., J Biomech 29 (1996) 1191-1194.

SBC2007-175825**MEASUREMENT OF ACOUSTIC EMISSION WAVE ATTENUATION BY BONES AND MUSCLES****Benjamin Pruden, Ozan Akkus**Weldon School of Biomedical Engineering
Purdue University
West Lafayette, IN**INTRODUCTION**

Stress fractures occur in bones of athletes and soldiers due to the accumulation of microcracks [1]. Detection of precursor acoustic emissions (i.e. ultrasonic stress waves) resulting from microcrack activity may help predict failure onset before continuous physiological activity results in full-blown fracture. An acoustic emission wave generated from a microcrack in bone will be diminished by dispersion, mode separation, reflection, and viscous losses induced by the biological tissues (skin, muscle, fat) between the source and the transducer. While others have recorded waves emanating from unknown loci in human knee *in vivo* using acoustic emission method [2], there is no means to appreciate how far these waves can travel in the body. Several studies have characterized the ultrasound attenuation in bone [3] and muscle analog homogenates [4] in the frequency range above 300 kHz. On the other hand, acoustic emissions are prominent in the range of 20 kHz to 300 kHz. *The current study focused on identifying the attenuation of acoustic emission waves in bone and muscle tissues in a frequency range which is more relevant to acoustic emissions. This information is critical for predicting whether an emission of certain magnitude at the source can reach surface mounted sensors without being totally attenuated.*

MATERIALS AND METHODS

The general approach of the study was such that specimens of varying thicknesses were prepared from muscles and bones. Acoustic emission waves were pulsed through these tissues and the reduction in waveform amplitude as a function of increasing tissue thickness was measured. The slopes of amplitude thickness plots yielded the attenuation of emissions in units of dB/cm. Attenuation was recorded for wave propagation along the transverse axis (direction transverse to longer axis of bone's shaft) and the longitudinal axis of bone. For the

muscle tissue, only the transverse axis (axis perpendicular to myofibrils) was quantified.

Transverse bone samples were prepared from the mid-diaphyseal shaft of a fully mineralized bovine femur (24-months old) using a low-speed diamond blade saw (Isomet 1000, Buehler, Lake Bluff, IL). The cortical shell pieces were further machined in to 0.75"x1" rectangular slabs of varying thicknesses. The thickness of each piece was measured twice using a micrometer (SPI2000, Swiss Precision) and the average thickness was used in later calculations. The thicknesses across which the pulses traveled ranged from 1.0 to 7.3 mm. *Longitudinal bone samples* were machined from the contralateral femur of the same animal. These segments ranged 0.34 cm to 8.34 cm in length with about 0.25 cm increments.

Bovine round steak was utilized for assessing attenuation of waves in *muscle*. Thirty samples with different transverse thicknesses were cut, using a hand saw, from the muscle while the muscle was frozen. Pieces which contained visible amounts of fat were excluded from the analysis. The pieces had a height and width of about 1" while the thicknesses along which pulses sent ranged from 0.81 cm to 7.6 with about 0.2 cm increments. The thickness of each section was measured using a micrometer in half-thawed state and care was given not to squeeze the tissue during these measurements.

Acoustic emissions were generated and recorded using two piezoceramic sensors (R15, Physical Acoustics Corporation, Edison NJ), preamplifiers (Physical Acoustics Corporation, Edison NJ) and a dedicated AE system (MISTRAS 2001, Physical Acoustics Corporation, Edison NJ). Each transducer was placed on opposite surfaces of the test sample using vacuum grease. The transmitting sensor was provided with a 5 μ s square pulse train per second for 30 seconds. The amplitude of the pulse was set as 100 dB. The waves traveled through the tissue and the amplitude was collected by the

other R15 sensor. The slope of the attenuation lines were obtained by linear regression analysis and a coefficient of determination value (R^2) was deemed significant at the level of $p < 0.05$.

RESULTS

The reduction of wave amplitude with increasing bone and muscle thickness was linear (Figure 1 a-c). The amount of attenuation was calculated as the slope of the best fit line and expressed in terms of decibel degradation per centimeter of tissue. The data show that for every centimeter an acoustic emission travels in the transverse direction of bone, longitudinal direction of bone, and transverse direction of muscle, the amplitude will be lowered by 2.48dB, 2.52dB, and 2.70dB, respectively. Despite the material anisotropy of bone, the attenuation was comparable in longitudinal and transverse axes. Attenuation through muscle was comparable to bone.

DISCUSSION

Knowledge on these simple measures of attenuation for muscle and bone would be valuable in several regards: 1) estimation of whether a wave can reach the surface mounted sensor, and, 2) provide a more realistic placement of acoustic emission sensors during *in vivo* sensing application to cover a wider space continuum with smaller number of sensors. Our earlier investigation on bone samples undergoing fatigue has demonstrated that the amplitude of emissions released due to microcrack activity range from 40 dB to 80 dB [5]. The lower threshold is determined by the level below which extraneous noise becomes a concern. The higher values occur during about the final 5% of the fatigue life where sample stiffness decreases exponentially. Lower amplitude events are associated with initiation and growth of smaller cracks and they occur much earlier during fatigue process. The attenuation coefficient obtained in this study predict that a low amplitude crack, say 50 dB, occurring in the medial aspect of tibia (which is a common site of stress fractures) would propagate about 4 cm before being attenuated below the detectable threshold of 40 dB. Medial aspect of tibia is the best case scenario as it is subcutaneous and attenuation by muscle is not a concern. On the other hand, a signal sourcing from proximal femur would have to travel 1 cm in bone and 6 cm in muscle before reaching surface, and the current attenuation coefficients predict that events less than 60 dB will not likely be detectable by surface mounted sensors at a threshold detection level of 40 dB.

The current results will be improved by conducting attenuation measurements through fat and skin. Furthermore, the attenuation due to transmission losses which sources from acoustic impedance mismatch at tissue boundaries needs to be assessed as well. Still, the current results indicate that even low-level microcrack events can be detected by surface mounted sensors for subcutaneous bones such as the tibia or metatarsals, sites which comprise 90% of occupational stress fractures [1].

ACKNOWLEDGEMENTS

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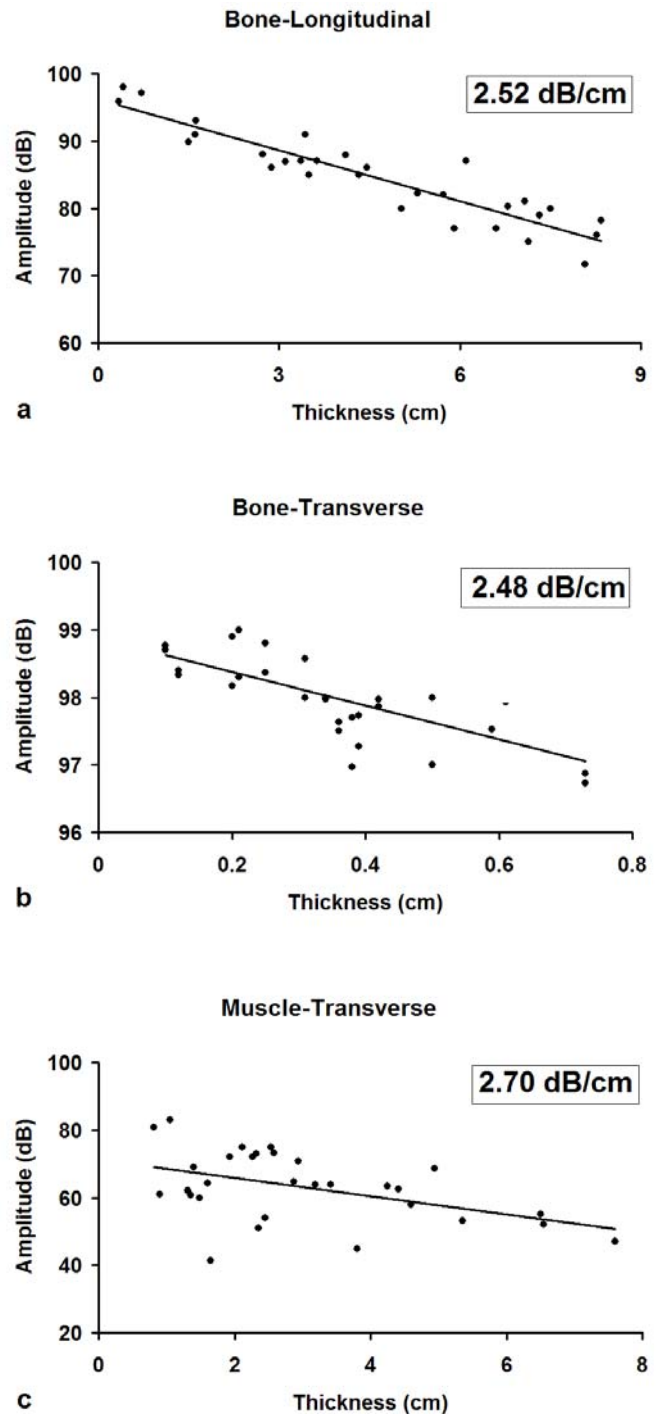


Figure 1. Amplitudes of transmitted acoustic emissions were obtained as a function of thickness for bovine bone. The best fit line was generated via linear regression and the slope of the line gives the attenuation coefficients which are denoted by the numbers in the boxes.

424. Detection Of Microearthquakes In Bone For Diagnosis Of Skeletal Fragility

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Most fractures of bone occur due to major trauma whereas much less appreciated are the spontaneous fractures which occur after accumulation of microtrauma over extended periods of time. Bones weakened by disease, disuse or aging are susceptible to such fractures in contexts of osteoporosis, diabetes, cerebral palsy, fibrous dysplasia and osteogenesis imperfecta. This study will demonstrate the feasibility of the acoustic emission (AE) method for in vivo noninvasive monitoring of damage accumulation to help these risk-defined populations by alerting them on an impending full-blown fracture. The two major requirements for AE's applicability in a diagnostic context are: a) emissions due to microcracks must have sufficient amplitude at the source, and, b) the source emission can reach the surface without being attenuated significantly by surrounding soft tissue. Emissions recorded from fracture of microcantilever bone beams (300x300 μm) have amplitude of 104.3 ± 2.6 dB at the source. Attenuation of surface born emissions along the spine and tibia were obtained in vivo by pulse transmission method as 0.5 dB/cm and 1.7 dB/cm, respectively. Attenuation of inborn emissions in dissected dog limb in vitro was more pronounced at 2.8 dB/cm. These results, with a detection threshold of 34 dB, predict the emissions from the body to travel in the range of 35 cm to 200 cm. Therefore, acoustic emissions due to skeletal microtrauma is likely to be detected in vivo provided that a portable emission system is fabricated. Design requirements of such a system will be elucidated.

Appendix V Could Acoustic Emission Waves Emanating from Bone Microcracks be Detected *In Vivo*?

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Introduction: Accumulation of microcracks in bones of athletes and military recruits leads to stress fractures [1]. These microcracks release elastic stress waves also called as acoustic emissions (AE). Stress fractures may be predicted by detecting such precursor AE events. As AE sensors are mounted on the skin, the knowledge on source intensity and attenuation of resulting waves while propagating between source and the sensor would be useful in predicting the distances that can be covered by AE. We shed some light on these questions via an in vitro micro-mechanical test model and in vivo pulse attenuation measurements.

Materials and Methods: Source strength Fifteen cantilever microbeams (cross sectional of 300 μm x 300 μm) were machined from bovine femur using a low speed saw (Buehler). These dimensions are within the range that is reported for in-vivo microcracks [2]. Beams were 3 mm in length and extended at the edge of bone wafers akin to a comb.

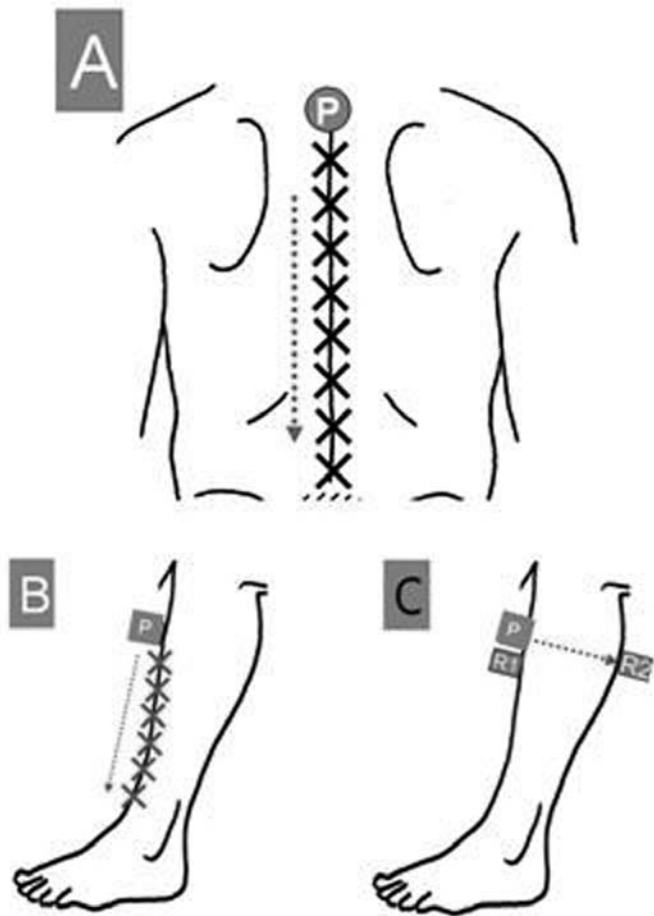


Fig. 1. Schematic representation of the attenuation measurement along the spine (A), along the tibia (B) and across the calf (C) (P: transmitter and receiver respectively)

Mechanical and acoustic emission testing: Microbeams were monotonically loaded to fracture at a rate of 0.15mm/sec by loading at their tips. The bone wafer to which microbeams cantilevered was mounted on translation stages which allowed positioning the loading tip at the same distance from the fixed end of beams. An AE sensor (R15, PAC) was mounted on the wafer at about a distance of 5 mm from the fixed end using high vacuum grease. Fracture load of each microbeam was collected and correlated with the amplitude of emissions resulting from the microfracture.

Attenuation: Two AE transducers (R15) of similar frequency response characteristics were used to assess attenuation of AE-like pulses along the length of a vol-

unteer's back (under IRB approval). The transmitting sensor (Fig. 1A, surface waves), was kept stationary and the receiving sensor is moved down along the spine. The transmitting sensor was pulsed and the amplitude of the AE was recorded at the receiver (AEDSP 32/16, PAC.). Similar procedure was carried out for the lower leg (Fig. 1B, surface waves). Waves were also introduced across the calf (Fig. 1C, through-pulsing). Attenuation for Fig. 1A and B were calculated from the slope of the line fitted to amplitude-distance plot (dB/cm) and through pulsing attenuation was calculated by dividing the difference between the source and receiver amplitude with the thickness of the calf.

Results: Beams failed with the release of a single emission. The intensity at the source was found to be 104.5 dB $\pm$ 2.8 dB (1641mV $\pm$ 526.2 mV in corresponding signal intensity). A significant correlation ($p < 0.05$) was found between AE intensity and fracture load (Fig. 2). The attenuation coefficient for surface waves in spine and tibia were 0.55dB/cm and 1.75dB/cm, respectively. For a detection threshold of 30 dB and an AE event of 100 dB source intensity, it follows that the wave can travel up to 127 and 40 cms on the surfaces of the back and the lower leg, respectively. In the case of through-pulsing the estimated travel distance before falling below the detection threshold is about 60 cm.

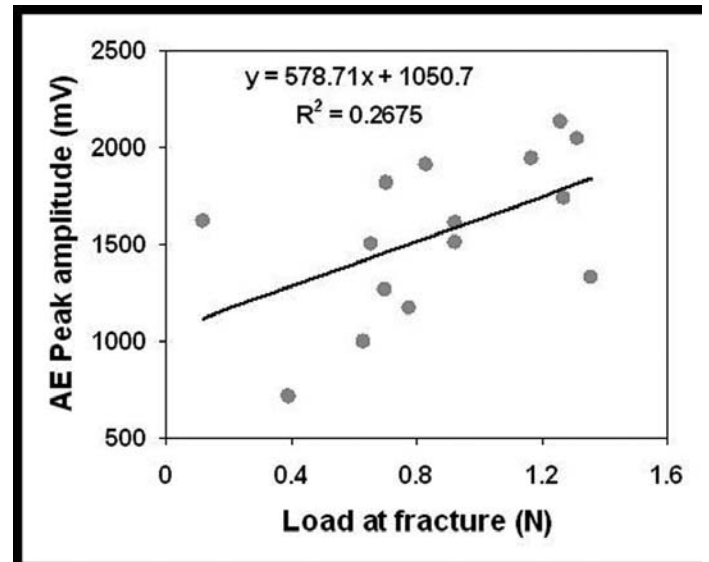


Fig. 2. Peak amplitude (mV) vs. load at fracture (N) ($p < 0.05$ for R^2)

Discussion: The results indicate that microcrack-like events can travel tens of cms in the body, implying that waves from in vivo cracks may potentially be recorded with a portable system. A basic appreciation of source intensity and attenuation along the path of an elastic wave allows: a) determining whether a signal can reach the sensor, b) finding minimum number of sensor for optimal coverage and c) improved localization of AE events. Currently we cannot explain as to why the attenuation of surface waves in tibia was greater than that of the spine. Measurements from greater number of participants and wave propagation simulations in these different settings may explain this variation. Ultrasound attenuation in bone and muscle homogenates has been traditionally studied in the MHz range [3,4]. Since the bulk of acoustic emission power spectrum lies in the lower range of 50–500 KHz, the current study reports attenuation in a more relevant frequency range. Acoustic losses due to fat, skin, muscle, and interfacial losses due acoustic impedance mismatch are currently being evaluated. In conclusion, results are supportive of the feasibility of AE based detection of stress fractures.

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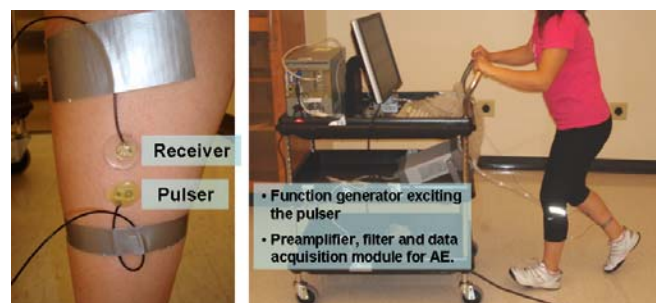
SBC2008-192778**FEASIBILITY OF PORTABLE ACOUSTIC EMISSION APPROACH FOR PROGNOSIS OF STRESS FRACTURES****Serife Agcaoglu, Vikas Kaul, Ozan Akkus**Weldon School of Biomedical Engineering
Purdue University
West Lafayette, IN**INTRODUCTION**

Repetitive mechanical loading induces microscale damage in bone to accumulate and may lead to stress fractures [1]. People with weakened bones due to disuse or disease, or, healthy people who have excessive exercise regimes (soldiers and athletes) experience these fractures [2]. Stress fractures interrupt training, reduce fitness and may even lead to discharge from the military in certain occasions [3]. Therefore, early prognosis and prevention of stress fractures would be desirable. Currently, following methods are being used for diagnosis: plain radiography, computed tomography (CT), bone scintigraphy and magnetic resonance imaging (MRI). The sensitivity of plain radiography is very low, 15-35% [4]. CT is less sensitive than radiography except some very special and rare cases of stress fractures [5]. Among these diagnostic methods, scintigraphy and MRI are more sensitive. However, the former lacks specificity because it may confound infections, tumors, bone infarctions, periostitis and osteonecrosis [2,5]. Furthermore, it is radioactive [5]. MRI has immense economical and logistical limitations [6].

Acoustic emission analysis can be a viable alternative for real-time, portable and non-invasive monitoring of the progression of stress fractures. Since it is known that accumulation of microcracks is the main reason for stress fractures [1], analyzing the acoustic emissions released by microcrack activity may be an efficient prognostic strategy for predicting failure of bone. However, an important detail to verify is whether or not there are extraneous emissions from joints or heel strikes which may confound with microcrack related emissions. In this study, we assessed whether there are artifactual emissions due to regular gait/running, and consequently, assessed the feasibility of acoustic emission approach for portable prognosis of stress fractures.

MATERIALS AND METHODS

The experimental setup consisted of two custom-made acoustic emission sensors, a function generator (Agilent 33220A) and a commercial acoustic emission system (Mistras, PAC, NJ) (Figure 1). The sensors have a layered design which consists of a PZT element between two epoxy layers. PZT gives the sensors the ability to detect high frequency AE signals and it is the sensing material in most AE sensors. The sensors were adhered to the tibia of the subjects (conducted under IRB approval) by a medical grade silicon agent (UroBond) within 15 mm distance of each other. One of the sensors acted as a pulser and was connected to the function generator to simulate acoustic emission signals. The pulser acted as a controlled input to verify the working ability of the receiver during the experiment. A sine-burst with a frequency of 200 kHz was supplied every 10 seconds, a frequency value within the frequency range for acoustic emissions. The other sensor, i.e. the receiver, was connected to the acoustic emission data acquisition system to record emissions from the pulser and other extraneous sources, if present.

**Figure 1. Experimental setup**

Electronics were loaded on a cart and the cables from sensors were connected. Power was provided by a long cable which allowed mobility for a range of 60 feet. The experiment consisted of two parts: running and jumping. Each part was conducted on three healthy subjects (24-36 years-old). Before conducting these experiments, the electrical noise floor was measured as 24 dB and the threshold was set to 30 dB accordingly. The first part of the experiment took 5 minutes for each volunteer and each ran a distance of 600 feet, while acoustic emission events were recorded. In the second part of the experiment, subjects jumped 60 times consecutively at a fixed location. For each experiment, the acoustic emission events were counted and recorded.

RESULTS

The total number of emissions during the running experiment for three subjects was 5.0 ± 3.0 . For an estimated number of steps of 400 over 5 minutes of running, the corresponding number of emissions per step was 0.0125. While jumping, 3.3 ± 1.5 emissions were received which corresponds to 0.055 emissions per jump. To evaluate the intensity of these emissions, we looked at the amplitudes. For running, the mean value for the amplitude of events was 36.5 ± 5.4 dB and that for jumping was 34.4 ± 3.1 dB.

DISCUSSION

In fatigue loading of cortical bone beams in vitro, failure takes place only after the release of hundreds of emissions (Figure 2). In the context of this abundance, the number of emissions due to artifactual emissions recorded during physical activity was negligible. Furthermore, these extraneous emissions fell in a narrow amplitude range. This increased our contention that these emissions were because of either a unique source or similar sources. It was also observed that the disturbing emissions coincided with unintentional pulling of sensor cables or the cables hitting the cart. One possible way to avoid these extraneous emissions is to increase the threshold since they fell into a narrow and low amplitude range (less than 40 dB). Our earlier experience of legitimate emissions from microcracks in cortical bone samples indicates a source amplitude in the range of 45 dB to in excess of 100 dB [7, 8]. For instance, had the threshold been set at 42 dB, we would not have recorded any emission artifactual emissions. The most ideal way to tackle the emissions due to cables is to utilize wireless sensors and receivers.

Frequency range of emissions is far greater than the frequency range of small deformations due to gait. This disparity explains the lack of emissions associated with heel strike or from joints themselves. Specifically, AE frequency range is greater than 100 kHz. A study by Fritton et al. investigated the small strains in bones of sheep and turkey *in vivo* during walking and it was demonstrated that the frequency content of deformations were within 40 Hz. Therefore, signals generated due to gait were very low compared to AE signals from microcracks [9].

In conclusion, artifactual acoustic emissions from young healthy subjects are minimal during physical activity. Therefore, acoustic emissions from microcracks occurring during prolonged strenuous activities can be evaluated without a significant burden of filtering noise. Currently, we are constructing wearable-pocket size devices to count emissions from subjects training in the field.

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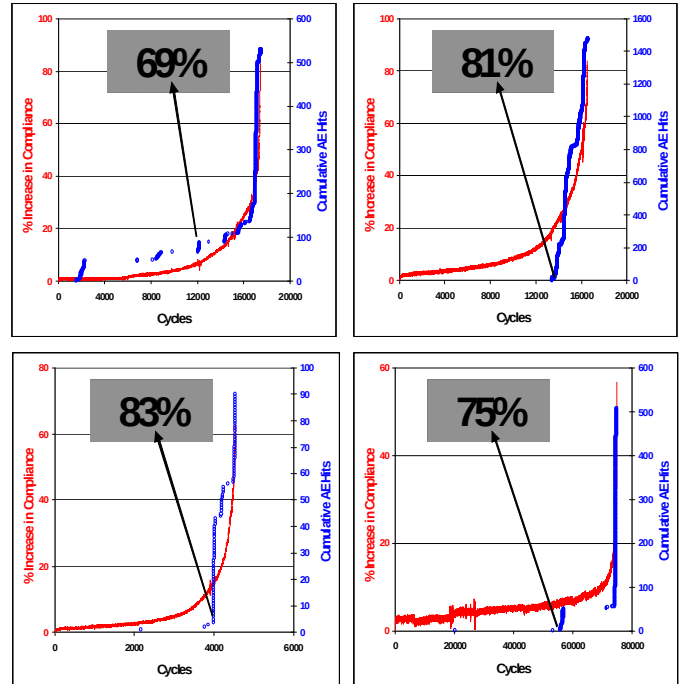


Figure 2. The change in compliance (red) and cumulative number of AE events (blue) during fatigue loading of cortical bone beams.

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