

**LASER INDUCED AUTO-FLUORESCENCE (LIAF) AS A METHOD FOR ASSESSING SKIN STIFFNESS  
 PRECEDING A DIABETIC ULCER FORMATION**

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**INTRODUCTION**

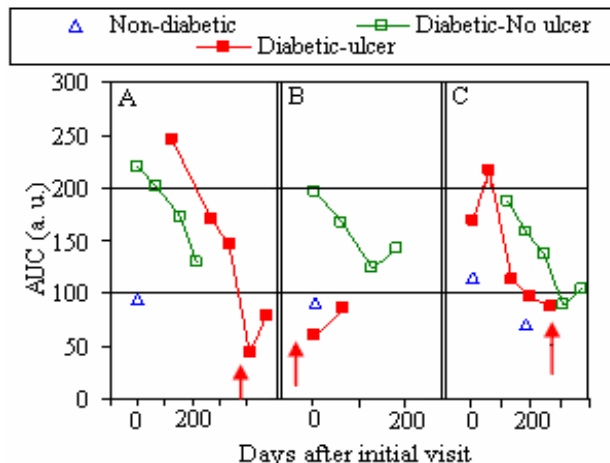
Diabetic foot ulceration is a leading cause of non-traumatic amputations. Discrepancy between the stiffness of the skin and the plantar soft tissues may influence the likelihood of ulceration. Changes in tissue properties with diabetes are mainly due to accumulation of glucose in tissues which promotes non-enzymatic glycation of structural proteins [1] such as collagen and keratin. Such intermolecular cross linking will alter the structure of the fibers and lead to their stiffening [2]. Our objective was to develop a non-invasive method for assessing skin properties in the context of ulcer formation on plantar surface of the foot in diabetic patients.

**METHODS**

A nitrogen laser (Laser Science VSL-337 ND) with excitation wavelength at 337 nm was used to induce fluorescence of the plantar skin of hallux and fifth metatarsal head from 9 male subjects (age 70-82, Table 1). The auto-fluorescence spectra were collected from each site with a fiber optic probe using over 500 diodes in the detector, corresponding to wavelengths of 300 to 650 nm. The spectral area under the curve (AUC) was calculated in arbitrary units after background subtraction and normalization. The diabetic subjects were monitored more frequently than the control subjects. Data was not collected from callused or open wound areas. Two-way ANOVA and student's t-test were used for statistical testing as appropriate.

**RESULTS AND DISCUSSION**

The two-way ANOVA of AUC indicated a significant difference among the three groups, but not due to skin sites. The AUC is significantly ( $p < 0.05$ ) higher for diabetic than in the non-diabetic individuals (Table 1). No significant difference was observed between the AUC of diabetic individuals based on ulcer formation. However most of those who developed an ulcer appear to have a sharp decrease in AUC about when the ulcer was detected (Figure 1). Only in one subject, the lowest point of AUC may have been missed since he started in the study while healing from an ulcer.



**Figure 1: Spectral characteristics of the plantar skin area of interest in age-matched subject groups based on time from their initial visit. AUC=area under the curve, au=Arbitrary Units. Arrows indicate ulcer.**

**CONCLUSIONS**

The LIAF data indicates a higher signal for the diabetic individuals due to their altered state of intermolecular bonds. However, the AUC decreased prior to an ulcer formation suggesting its potential as a marker of tissue stiffness and thickness changes which precede ulceration in the diabetic foot. Additional data are needed to evaluate the differences within the diabetic population which predispose some to ulceration.

**REFERENCES**

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2. Monnier, VM, *et al. The New England Journal of Medicine* **314**(7):403-408, 1986

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**Table 1:** The AUC of LIAF spectra and age of the subjects (n=9). Groups A, B, and C correspond to Figure 1 based on ulcer site.

| Max.AUC (arbitrary unit)   | Group (Fig 1) | Non-Diabetic | Diabetic-No Ulcer | Diabetic-Ulcer |
|----------------------------|---------------|--------------|-------------------|----------------|
| Hallux                     |               | 130.7        | 196.9             | 126.7          |
|                            | A             | 95.4         | 220.0             | 245.3          |
|                            |               | 98.3         | 178.0             | 92.4           |
| 5 <sup>th</sup> Metatarsal |               | 96.8         | 205.4             | 202.0          |
|                            | B             | 91.3         | 198.5             | 85.8           |
|                            | C             | 115.0        | 187.4             | 217.6          |
| Age (Years)                |               | 76.7 ± 4.6   | 76.3 ± 3.2        | 73.7 ± 5.5     |