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ESPI and PulsESPI Applied to Ophthalmology Using Modified Twymann-Green Interferometer

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Abstract: Perhaps for the first time ever, application of Pulsed Electronic Speckle Pattern Interferometry (PulsESPI) technique to an animal (bovine) cornea for the investigation of its dynamic behavior to localized stresses is being presented. For the target of obtaining optimal results from laser ophthalmic surgery for the correction of cornea, it has already been observed that more attention is required on the adjustment of the necessary ablative laser power, while taking into account the otherwise neglected corneal elasticity as a parameter. It is being proposed that this need can be fulfilled by analyzing through this technique the behaviour of cornea under application of variable dynamic stresses. By addition of elasticity to the related cornea mathematical model, corneal healing evolution can also be forecasted for follow-up and for perfecting the correction during the post-op period. The tests were complemented by an *in vitro* measurement of the surface contour of the same cornea using a Twymann-Green Interferometer.

Key words: ESPI, PulsESPI, interferometer, cornea, stress

INTRODUCTION

Corneal corrective laser surgery procedures like PTK and LASIK have made dramatic improvements in the last decade, yet a large number of patients cannot achieve targeted correction. Many machines, especially of early generations, are excessively rigid, not allowing adaptation to the intricacies of the corneal changes (Gualini *et al*., 2000). Use of corneal topographic devices like videokeratometers has proven insufficient since these devices operate at resolution of around 100 microns, while ablation depth is less than few microns in total and single-shot removal is in sub-micron range. White light interferometers have better resolution of around 10 micron but it is still not sufficient.

In fact, a wave-front analysis does not give any information about the critical parameter of corneal elasticity. But the corneal surface reconstruction process after ablation depends greatly on the corneal tissue elasticity, as confirmed by the results obtained by using steroids to modify the intra-ocular pressure or IOP during the post-op period. Thus a real improvement in PTK procedures would be to combine wave-front analysis with an effective method that takes tissue elasticity into consideration (Drescher *et al*., 1999; Kasprzak *et al*., 1994; Matsuda *et al*., 1982). The use of a Twymann-Green Interferometer (TGI) (Gualini *et al*., 2000; Licznerski *et al*., 1999) for mapping of corneal surfaces and to also measure their variations due to intrinsic elasticity coefficients is proposed. Such system can go to sub-micron resolution and should prove to be an attractive alternative.

For mapping the behaviour of cornea under dynamic stresses, a very innovative laser device, i.e., a PulsESPI camera, is being used. PulsESPI is very good candidate for this purpose as its resolution goes to sub-micron ranges. Through this study, the resonance frequency and the corresponding corneal deformations are determined in real time, probably for the first time.

MATERIALS AND METHODS

An experiment was conceived in order to evaluate the use of a PulsESPI camera to measure the animal corneal deformations in real time. This research should pave way to *in vivo* experiments on human cornea. A normal bovine cornea was used for the study, which is probably not the best choice to emulate a human cornea, since other smaller animals (e.g., goat, sheep, pig) may be more comparable. Along with the reasons of availability and the drawback of having low reflection coefficient to visible

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light, it does offer quite a wide surface so that fringes can be conveniently displayed and the tip of a shaker could easily be applied without significantly disturbing the field of view. The experiments were conducted at Steinbichler Optotechnik GmbH in Neubeuren, Germany few years back. The authors jointly took the responsibility of experimentation and analysis, which took few weeks for completion.

A PulsESPI device is being chosen for the study because of the many advantages offered by this technique in real-time deformation analysis. PulsESPI is becoming increasingly popular as a reliable and accurate investigation method compared to conventional Double Pulse Holography (Kasprzak *et al*., 1994; Bally, 1979; Friedlander *et al*., 1991; Ohzu and Kawara, 1982). Here, instead of using photographic films, PulsESPI can directly record images in the PC through a CCD camera. PulsESPI images can be obtained with very short light pulses at high repetition rates, which make the method insensitive to mechanical vibrations. Pulse widths and repetition rates can be adjusted conveniently to cover quite a wide range of applications. Due to the short recording time (few nanoseconds) PulsESPI is insensitive to disruptive factors like low frequency vibrations or shocks transmitted to the apparatus. Presently PulsESPI is probably the best investigational method to study transient and dynamic behaviour of objects under excitation. It largely combines the advantages of both conventional Double Pulsed Holography (DPH) and ESPI. Figure 1 shows a basic layout of a Twymann-Green Interferometer. Figure 2 shows the basic layout of a PulsESPI system.

In these experiments, data was acquired and processed by the powerful software FRAMESplus release 5.0, developed by Steinbichler Optotechnik GmBH. The equipment used for this experiment has a resolution ranging between λ/30 and λ/10 at 20 pixels fringe width. The camera resolution is 1280×1024 pixels. The ruby laser pulse separation ranges from 2 to 800 μsec. Polytec Laser Vibrometer was used to determine the resonance frequency.

For inducing strain to the bovine cornea a mechanical vibrator in the shape of a rod connected with a motor was used. When the motor was electrically driven, the tip of rod would poke the sample with its tip moving back and forth with a displacement of a fraction of millimeter. The oscillating frequency of the tip was variable over wide range and Polytec laser vibrometer was used to measure its frequency. The bovine cornea was supported by aluminum film and polystyrene.

In the PulsESPI experiment, the ruby laser light was widely diffused, thus the back-scattered signal was found to be very poor due to the high transparency of the bovine cornea. This problem was overcome by spraying the corneal surface with white powder film in order to ensure high reflectivity and uniformity of the back-scattered signal. This may appear a limitation for an *in vivo* application on human cornea, but in that case a comparatively better reflectivity to the ruby laser (632.8 nm) is expected, as already reported by other authors (Kasprzak *et al*., 1994; Bally, 1979; Friedlander *et al*., 1991; Ohzu and Kawara, 1982).

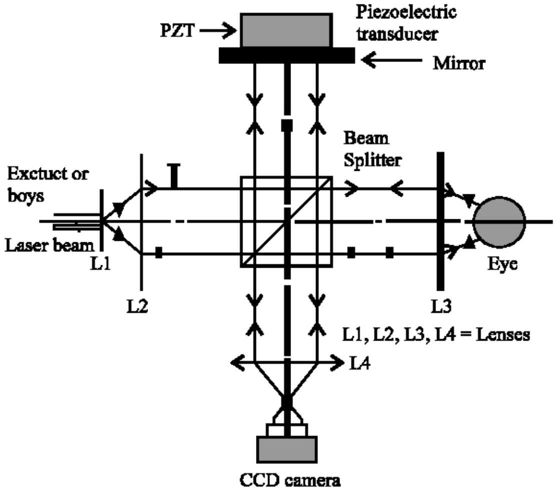

Fig. 1: Layout of a basic TGI setup

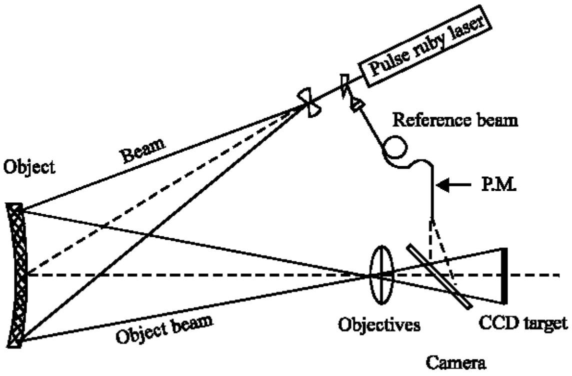

Fig. 2: Layout of a basic PulsESPI setup

RESULTS

Initially, some static measurements were performed using a TGI setup in order to map the static condition of bovine cornea. The setup of the TGI experiment is visible in Fig. 3, while Fig. 4 shows the bovine cornea surface mapped by the TGI fringes. Fringes generated on the

Fig. 3: Setup of the TGI experiment

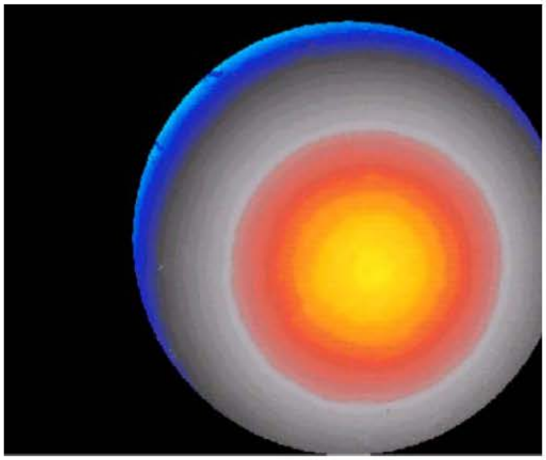

Fig. 4: Bovine cornea mapped with TGI

surface of the bovine cornea were processed by FRAMESplus release 5.0/1, which generates results within few seconds (Licznerski *et al*., 1999).

Once the cornea was mapped without deformation, a specific static stress (controllable in intensity and direction) was induced to the bovine corneal surface as visible in Fig. 5. The resulting corneal deformation mapped by the TGI device and obtained after the fringe analysis from the software is shown in Fig. 6.

Preliminarily, a HeNe laser is used to align the optical system in eye safe conditions for the operator, before the actual use of the high power ruby laser. Part of the beam is injected into a single-mode, polarization-maintaining optical fiber, which carries the reference beam signal directly onto the CCD camera surface.

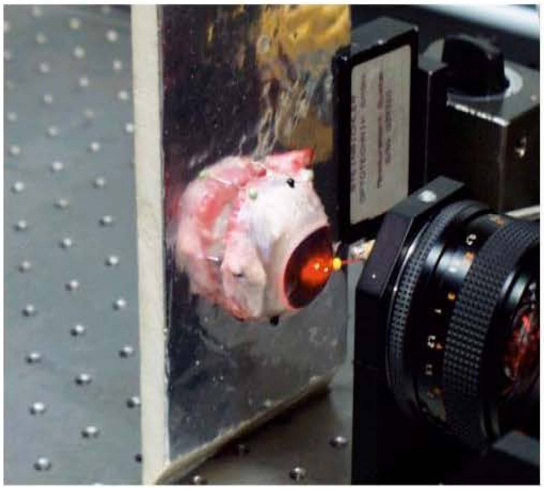

Fig. 5: Sample fitted in TGI experiment

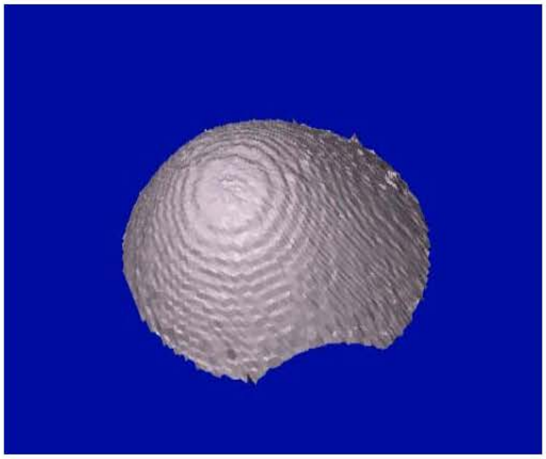

Fig. 6: Static deformation mapped with TGI

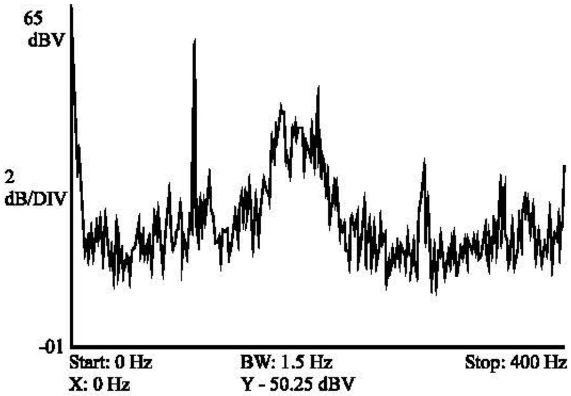

Fig. 7: Vibration frequency spectrum for the cornea

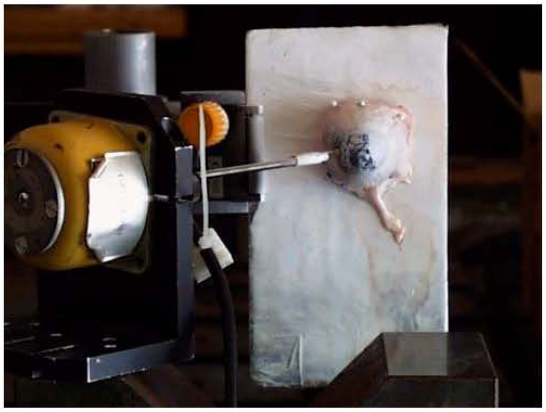

Fig. 8: Dynamic stressing setup for the cornea

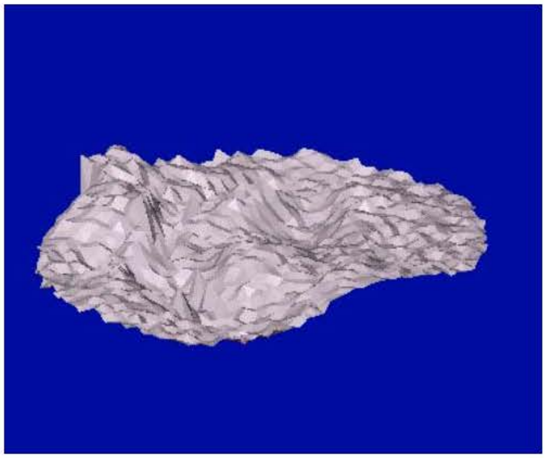

Fig. 9: Deformation of cornea at resonance

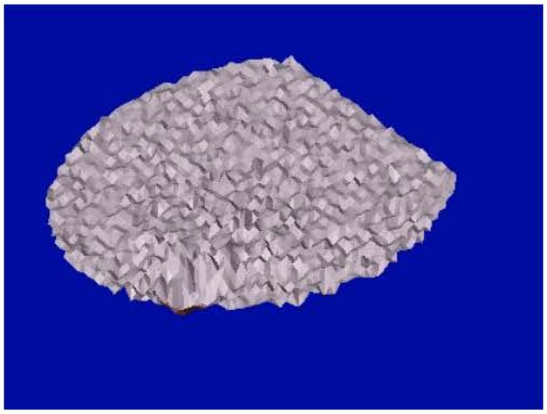

Fig. 10: No deformation of cornea outside resonance

Figure 7 shows the amplitude response versus the frequency spectrum of the bovine cornea surface shacked with the tip synchronized to the Laser Vibrometer. Figure 8 shows the measurement setup. The graph of the amplitude versus frequency spectrum is directly obtained from the vibrometer. The resonance frequency is clearly visible at 175 Hz. Thus, the tip vibration frequency was fixed at resonance conditions and the surface deformation at this resonance frequency was detected using PulsESPI setup. As is visible from Fig. 9, the cornea looks evidently deformed. Quite interestingly there are no apparent measurable deformations at frequencies slightly offset from the resonance, as visible in Fig. 10. The bumpy surface is due to unfiltered calculation noise generated by the software, which is shown in the original form.

DISCUSSION

Firstly, the fast and direct but static mapping of cornea with the help of modified Twymann-Green Interferometer were obtained. Though it gives impressive output, the target here was to obtain the mapping of the cornea under dynamic stress applications which is necessary to obtain the elastic behaviour of the sample. PulsESPI was able to provide this important information to us. This technique can be used for the systematic studies and investigations on the elastic behaviour of cornea. Information of the cornea elasticity can be very well utilized for fine-tuning the final surgical ablation procedure.

It was found out that the cornea surface as used in this experiment exhibits a resonance frequency which can very well give information about its elastic parameters. This information of having a specific resonance frequency can be characteristic to a specific cornea or the eye. Also evident is that PulsESPI is a dependable and promising method for the conduction of stress-related investigations on a biological sample (Reiss, 2003; Tyrer, 2001).

Looking at the earlier researches for the development of topographers and videokeratormaters, it is found that the corneal elasticity is generally neglected parameter in these studies. Using PulsESPI to extend the studies in this direction opens up another avenue which will pave way for other stress-related studies on biological samples with this technique.

Preliminary studies on animals can be useful to determine all the process parameters and functions needed for elasticity determination. As an implementation, treating patients with excimer laser and then monitoring the corneal evolution during the post-op time can be compared with the calculated figures of forecasted cornea surface. This procedure will enable us to refine a mathematical model so that it can be used *in vivo* conditions for humans (Garcia *et al.*, 1998, 1999).

CONCLUSIONS

It is suggested that normal optical methods like TGI or wave front analysis are not sufficient for completely establishing the Targeted Correction, unless corneal elastic properties and coefficients are duly taken into account. Thus stress analysis should help to predict the post-op time evolution of the corneal tissue.

In an attempt to demonstrate findings, the bovine cornea was mapped at rest and under static and dynamic stress conditions. The optical measurement methods are used that involve data acquisition without contact with the sample.

To map the cornea at rest and under static stress conditions, a modified Twymann-Green interferometer is used. Then for the first time ever PulsESPI is utilized to measure and map the deformations of the bovine cornea to a dynamic stress and to find the intrinsic resonance frequency of the bovine cornea. It was finally noticed that outside resonance condition the bovine cornea is practically not deformed.

The technique can then be adapted and developed for *in vivo* experiments on animals and humans. This would yield a very accurate definition of corneal mathematical model, so that a prediction curve of the cornea evolution with time can be visualized. It is also proposed that PulsESPI can be used in combination with a wave front analysis device to achieve much better results compared to their isolated use.

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