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Development of a Novel Micromechanical Tester for Biomedical Engineering Applications

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INTRODUCTION

The focus of this report is on the development of a micromechanical tester that can be used to study micrometer thick biomaterials and biological tissues. The tester can be mounted onto an X-Y stage of an inverted or compound microscope to observe the microscopic deformation of the test specimen during testing.

The study of the mechanical properties of materials is important in the design and fabrication of any microscale product. Acquiring information such as the fracture toughness, fatigue limits, ultimate tensile and yield strength of these materials would help to determine the reliability of the final product made using the material.

Traditionally, these material properties are obtained via mechanical testing on a macroscale tester such as the machines produced by Instron. However, mechanical testing of ‘softer’ materials with a microscopic size is more complicated as the test procedures and equipment have to address concerns such as clamping and alignment of the specimen. Microscale mechanical testing is a new area of study that tests specimen of miniscule dimensions. Thus, knowledge on microlevel testing is still limited. More accurate and reliable approaches are needed in the development of the techniques in micromechanical testing.

In the early 2000s, horizontal micromechanical testers became commercially available. These testers primarily utilize the concept of a power screw system to provide a controlled translation to generate axial displacement on the specimen. Deben UK (2011) (http://deben.co.uk) is one of the well-known leaders in the design and production of micromechanical testers, covering both horizontal and vertical testing. Their horizontal testers are mainly intended for mounting specimens aligned on the loading axis. These rigs can be mounted onto a conventional microscope for specimens to be viewed, but are incapable of being used on an inverted microscope which allows for certain techniques such as the differential interference contrast (DIC) microscopy, phase contrast optics and Kohler Illumination, which are more superior for microscopic viewing of biomaterials, livings cells and translucent specimens. Also, these rigs are not cheap; they can cost up to SGD$25,000 per set.

Fig 1. The Purslow micromechanical tester.

We fabricated our first in-house micromechanical tester in 2008, based entirely on Professor Peter P Purslow’s approach and previous Purslow prototypes (Fig. 1). The tester has been used extensively to study micrometer thick tendons from the tail of rodents (Goh et al. 2008; Goh et al. 2012; Goh et al. 2010; Goh et al. 2014; Yeo et al. 2011), silk fibres (Lai & Goh 2015), micrometer thick polymeric membranes (De Silva et al. 2013) and biopolymeric fibres (Xie et al. 2008; Chew et al. 2011; Wang et al. 2015; Wang et al. 2016). It was a low-cost alternative to the commercially available ones. The tester housed a large cylindrical direct current (DC) motor to drive one of the pair grips while the other grip was stationary as it was attached to a load cell. The DC motor shaft was coupled to the grip by a Kevlar string; the rotation of the shaft in one direction then caused the string to coil around the motor shaft and this produced a pulling action on the grip. When a specimen was mounted between the grips, this action then resulted in a tensile force on the specimen and the specimen deformed in tension as intended. A compact linear variable differential transformer (LVDT) provided the positional measurement.

A petri-dish could be installed in the test chamber (secured by screws) to enable continuous hydration of the specimen if needed. The main issue with this tester was the drive system: (1) slacks in the Kevlar string could result in artefacts in the load-displacement data and (2) the DC motor was bulky and stood in the way of the lens from a compound microscope.

With regards to the above limitations, clearly, the drive system of the in-house tester would have to be redesigned. Here we discussed the design and development of the Mark 2 prototype of the micromechanical tester, the experiments that we have carried out to use the prototype to evaluate the mechanical properties of microspecimens and the results arising from these experiments.

MATERIALS AND METHOD

Core design specification

From the perspective of performance, the micromechanical tester should have the following features: a capacity to test specimens to loads of less than 20 N, a linear scale resolution of approximately 0.067mm/s, able to secure petri dish (commercially available 60-mm diameter ones), can be mounted on an inverted or compound microscope x-y stage, comes with removable specimen grips (that facilitates specimens to be mounted to the grips before the grips are introduced into the tester), a wide field of view under microscopic observation during testing, single-operator ease of operation, and computer control during testing and data acquisition.

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From the perspective of the test environment, the temperature during testing is expected to be room temperature in a laboratory environment. One may also expect that the specimen may require exposure to both acidic/alkaline solvents during testing.

From the perspective of maintenance, one expects that the operator would perform periodical washing of components to remove saline exposure (if grips and specimens are immersed in the petri-dish containing the saline solution during testing), calibration of the tester, and checks on the actuator and load cell.

From the perspective of the size of the tester, one expects that the tester should be able to fit onto the microscope X-Y stage which is typically about 170mm x 225mm x 190mm. The tester should be relatively portable, of about 3 kg.

From the perspective of material selection for the tester, one should expect that the material is resistant to humidity, alkaline and acidic environment. Additionally, the grips and related structures would have to be able to resisting yielding during testing and the grip system should not flex under load while in operation.

Prototype

The schematic of the assembly of the Mark 2 tester is shown in Fig. 2. The Mark 2 prototype is shown in Fig. 3. The rig was dimensioned to accommodate the selected actuator and load cell with clearance slots incorporated to house their wires without distorting or bending them. The actuator selected for the prototype was purchased from Zaber Technologies Inc. (model T-NA03A25; www.zaber.com); the load cell was purchased from Interfaceforce (model WMC-22N; www.interfaceforce.com); the LVDT was from TML (model CPD-M, www.tml.jp). An alternative load cell, UF1 (Low Range Isometric Force Sensor, LCM Systems; maximum 450 g) could be adapted to this system if needed. The computer control of the actuator involved software provided by Zaber; the acquisition of loads from the load cell was handled by a low-cost computer data logger (Pico Technology, www.picotech.com).

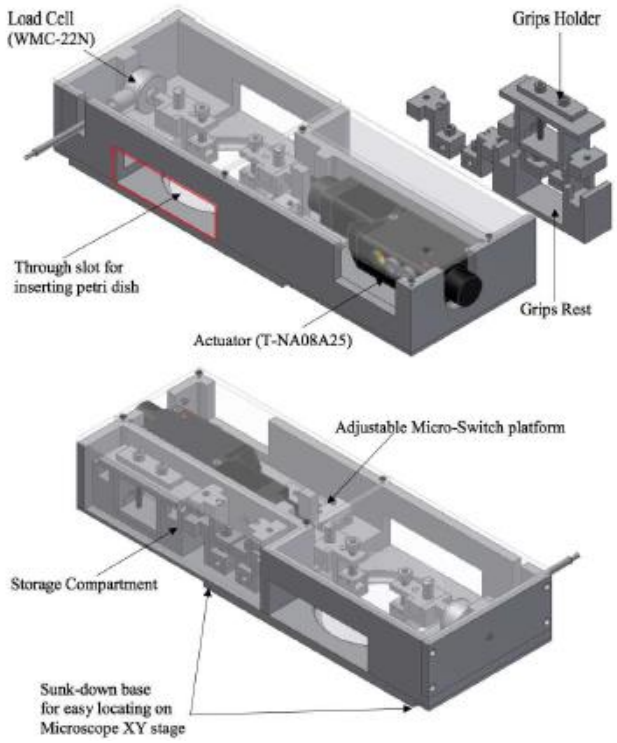

Fig. 2 Schematic of the Mark 2 micromechanical tester.

For ease of installing the petri-dish onto the tester, a slot was created at the side of the frame of the tester to allow the dish to be inserted into the test chamber.

The base of the test rig was designed with a sunken feature to enable the tester to be 'locked' onto the X-Y table of the microscope stage. The petri-dish pit was designed with a sunken feature to secure the Petri dish as well as to create sufficient working distance between the specimen (when mounted onto the grips) and the microscope lens.

A storage compartment was incorporated to house all the accessories pertaining to the grips. Of note, the grips featured simple plates for sandwiching the ends of a test specimen.

The grip-holder and grip-rest were accessories for supporting the manipulation of the grips and specimen outside of the tester. The griprest serves as an external support structure for securing the specimen outside the tester; the grip-holder serves to facilitate the transporting of the specimen into the test rig.

Aluminium alloy was used for the tester frame (main body) and internal structures because of its inherent resistance towards various environmental conditions, light weight, and high machinability.

Aluminium alloy was also used in the manufacture of the grips. All in all, the total cost of the fabrication (including the material) of the tester, the linear actuator, the load cells, the LVDT, and data logger was less than SGD5000. This was five times cheaper than a commercially available equivalent tester.

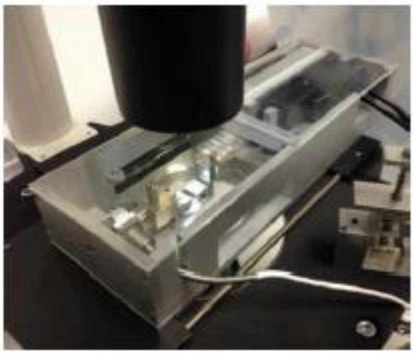

Fig. 3 The Mark 2 micromechanical tester. The prototype is shown mounted onto the stage of an inverted microscope to provide real-time examination of the specimen deformation during testing.

Mterial testing

The Mark 2 tester has been used to evaluate the mechanical properties of single fibres from three types of natural fibres, namely yarn flax fibres (Fong et al. 2015), oil palm empty fruit bunch fibres (Tan et al. 2017) and coir fibres (Thomas et al. 2017). Each fibre featured thickness on the order of micrometer. Of note, these fibres were chosen because they are well-regarded for their applications in medical sutures and composites. These fibres were obtained from the local plantations in Kuala Lumpur and Johor Bahru (Malaysia). In the next section, we highlight the performance of the tester by briefly discussing the results obtained from these fibres.

RESULTS AND DISCUSSION

Application 1: The mechanical properties of yarn flax fibres for polymer composite sutures

Flax (Linumusitatissimum) is a blue-flowered herbaceous plant that is found in temperate zones. The plant can be cultivated for its fibres and oil. From biomedical engineering, an important application of flax fibres is in surgical sutures (Linatrix@, B. Braun Sutures, www.aesculap.de). In this study, we were interested in the mechanics of flax fibres with a focus on the effects of moisture on the mechanical properties of yarn flax fibres as well as the possible dependence on knot geometry (Fong et al. 2015). Further details concerning this study can be found elsewhere (Fong et al. 2015). In the study, by testing the flax fibre immersed in water in the petri-dish, it was found that moisture in the yarn flax fibre has a significant influence on the mechanical performance of the fibres. Dry yarn flax fibres exhibited brittle fracture; wet yarn flax fibres showed clear features of fibre debonding, leading to fibre pullout (this finding was derived from observation under the microscope during testing). When knotted fibres were tested, all (dry and wet) fibres ruptured at the entrance to the knot. However, under the microscope it can be seen that the fracture morphology at the ruptured ends from dry ones exhibited brittle fracture but for the wet ones, fibre debonding

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followed by pull out were observed. These variations in the fracture morphology implicate the underpinning different mechanics of the wet and dry yarn flax fibres for taking up loads. Evaluation of the stress-strain data to derive the stiffness and strength of these fibres showed that dry fibres were stiffer and stronger than the wet ones (although the differences were not appreciably large). However, the wet fibres were more extensible than the dry ones. Overall, these findings were found to be useful for use in the design of polymer treated flax fibre for, e.g., sutures as well as hand layup polymer composite laminates.

Application 2: The mechanical properties of oil palm empty fruit bunch fibres for biopolymer composites

Oil palm (Elaeis guineensis) is a tropical tree. This tree is widely cultivated for the mesocarp (reddish pulp), a part of the fruit of the tree because the mesocarp can be processed to yield the edible vegetable oil which is a high-demand global commodity. The processing of the mesocarp can result in a lot of biowaste such as oil palm empty fruit bunch (OPEFB). The OPEFB contains a lot of fibres which are currently lending to opportunities for use in reinforcing engineering materials. In this study we were interested in the mechanics of oil palm fibres with a focus on the effects of moisture on the mechanical properties of the fibres (Tan et al. 2017). Moisture in natural fibres such as OPEFB fibres can have a significant influence on the mechanical performance of the fibres. Although many studies have sought to test them in bundles, it is not entirely clear how the mechanics of these fibres in wet and dry state affects the bundle properties. Here we tested these fibres respectively in dry and wet (immersed in the Petri-dish) state using the micromechanical tester (Tan et al. 2017). We found that the dry fibres exhibited mode 1 and 2 fracture as observed under the microscope during testing. We also observed that moisture-treated OPEFB fibres exhibited mode 1 fracture as well as defibrillation, and subsequently, microfibril pull out occurred. These observations of the fracture morphology suggest that the wet and dry OPEFB fibres possess different mechanics for taking up the load. On evaluating the stress-strain data, we found that the dry fibres were 4 and 2 times, stiffer and stronger respectively than moisture-treated fibres. We found that there was not a significant difference in the extensibility of dry and moisture-treated fibres.

Using these results, we were able to establish arguments for designing OPEFB fibre reinforced biopolymer composites such as starch-based composites. For further details, see the report published elsewhere (Tan et al. 2017).

Application 3: The mechanical properties of single coir fibres

Coir fibres (from the husk of the nut) can be extracted from the husk. The extraction process produces bundles of coir fibres, and after subjecting to additional processing they may then be blended with polymer-based materials to produce a variety of composite materials, e.g. blending with natural rubber latex for the production of medical implants (Geethamma & Thomas 2005). The performance of the fibres when used in composite material for engineering applications, especially the capacity of the individual coir fibres for composite reinforcement under different environmental conditions, has been the subject of many studies. Several issues of interest are as follows: coir fibres deformation and rupture, fibre-matrix delamination, matrix ruptures around the fibre and fibre pull-out from the matrix, and, the performance of the fibres in the composite when water gets through the composite into the fibres. Of note, coir fibres are hydrophilic. In humid environments, moisture diffusion into the composite could occur; this could degrade the composite mechanical properties and performance. Here, we were interested in understanding the mechanics of the single coir fibre in a hydrated state (Thomas et al. 2017). Similarly, specimens of coir fibres were subjected to tensile test until rupture using the micromechanical tester, mounted on an inverted microscope. The hydrated specimens were submerged in water held in a Petri-dish for continuous hydration during the test.

Briefly, the measurement from micrographs showed that the fibre diameter of the wet specimens were larger than the dry specimens. We also found that there were strong evidence of differences in the following parameters, stiffness, and strength. We found that the dry specimens were stiffer and stronger than the wet specimens. However, we observed that there were no differences in the extensibility of the wet and dry fibres. For further details, see the report published elsewhere (Thomas et al. 2017).

CONCLUSION

This report has shown how an in-house developed micromechanical tester could be cheaply fabricated and used for simultaneous mechanical testing and microscopic visualisation of micrometer thick specimens. The tester was used to evaluate the mechanical properties of three common natural fibres, namely flax fibres, oil palm empty fruit bunch fibres and coir fibres. The unique ability of the tester to accommodate fibres in a horizontal position for mechanical testing has enabled (single) fibres to be tested under two conditions, dry and wet (continuous hydration) states. Although the examples provided here are fibre specimens, other materials such as very thin membranes may also be tested using the micromechanical tester.

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