# DIMENSION MEASUREMENTS FROM PICTURES OF TENSILE TEST SAMPLES: GUIDELINES TO IMPROVE REPRODUCIBILITY

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#### Introduction

Studies characterizing experimentally biological tissues abound in the scientific literature, but with a large variability on outcomes [1]. C4Bio (https://c4bio.eu/) is an international community challenge founded in 2021, in which two testing campaigns were conducted to define and evaluate a consensus methodology for characterizing biological and synthetic samples from tensile tests. The observed inter-laboratory variability may be partially attributed to the measurement of the sample's cross section. Measuring dimensions on soft tissue samples with a caliper is still a challenge, and optical method may be a good alternative. This study aims to improve the reproducibility of sample dimension measurements from pictures, as plan during the C4bio challenge, and to estimate the effect of their uncertainty on the stress-strain curves through uncertainty propagation.

## Methods

**Image acquisition.** Twelve synthetic samples were cut out from an industrial silicone plate using the C4Bio cutting tool (theoretical section area 4\*2mm<sup>2</sup>). Samples' pictures were taken with a high-resolution camera (COOLPIX P7100, ISO 100, 3648\*2736 pixels) and calibrated using a millimeter (mm) paper. The samples were aligned with the mm grid. Dimensions were measured using Fiji (v1.53t, https://fiji.sc/) from a top picture (Figure 1) for the width, and from a side picture using a C4Bio holding device for the thickness.



Figure 1: Top and side pictures of a tensile sample

**Initial procedure.** Images were calibrated with a polyline of 4 points spaced by 10mm. Sample dimensions were the average length measured between 2 points at 5 locations in its reduced section.

**Extended procedure.** Additional instructions were followed. Images were first filtered (local contrast enhancement, sharpen); the points should be pixel-accurate placed in a high contrast area, within the calibrated zone, and should be checked a posteriori.

**Reproducibility.** Three operators applied the two procedures. Scilab (v6.1.1 <u>https://www.scilab.org/</u>) was used for processing the dimensions and statistics (mean, standard deviation (SD), inter-quantile (IQ) range, Wilcoxon signed rank test...).

**Effect on the stress-strain curves.** The thickness and the width SD, plus a SD of 0.2N on the force, were propagated as independent variables on the force-strain curves from experimental tensile test of the samples.

## Results

The picture mean scale was (mean  $\pm$  2SD) 18.1  $\pm$  38 pixels/mm. The thickness and width distributions were non normal for the two procedures. The initial procedure led to the same variability for thickness and width (2.347  $\pm$  0.147 and 4.027  $\pm$  0.157mm respectively). Five out of 72 measures were identified as outliers. Two operators provided significant different sets of width measures (p<0.05). The extended procedure eliminated the risk of outlier and the significant difference between operators, and decreased the variability  $(2.293 \pm 0.124 \text{ and } 4.080)$  $\pm$  0.108mm). It led to statistically different measures from the initial procedure (p < 0.0003), the mean and median being in the same order. The width IQ was reduced (from 0.087 to 0.055mm), not the thickness one. For both the initial and extended procedures, the stress SD along the mean stress-strain curve (3.7% and 2.9%) remains within the experimental corridor (5.2%), the extended procedure slightly reducing the stress range.

## Discussion

Simple guidelines can help to reduce the uncertainty on dimensions. Filtering underlines the details of the picture and standardizes the image interpretation made by the operators; and a pixel-accurate location is closed to a 0.05mm uncertainty. But high-quality images are essential for the reliability [2]. On side pictures, due to the sample holding configuration, either the sample's side or the mm grid were blurred, rendering the selection of points less accurate. In spite of this, the uncertainty on the dimensions of the section was coherent with the literature in percentage [3] and did not increase the variability observed experimentally on the stress-strain curves. Thus, it seems that, reducing the intra-operator variability can reduce the geometrical uncertainty, but this variability could not explain the inter-laboratory variability observed on the stress-strain curves [1].

## References

- 1. Singh and Chanda, Biomed Mater, 16(6), 2021.
- 2. Yang et al, Scientific reports, 10(1):1-8, 2020.
- 3. El Fazani et al, Measurement, 202, 111726, 2022.

## Acknowledgements

We thank the teams involved in the C4Bio Grand Challenge.

