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Independent Study on the stiffness properties of pure PDMS elastomers and stiffness tuning of magnetorheological PDMS elastomers

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Abstract:

The aim of this independent study is to find a way to maximize the change in Elastic Modulus (stiffness) induced by applying magnetic fields to magnetorheological elastomer (MRE) devices made by combining Polydimethylsiloxane (PDMS) elastomers with metal particles. In order to achieve this, I tried changing conditions that I expected to impact the elastic modulus change induced by applying magnets to MREs, including MRE thickness, composition (wt% of carbonyl iron particles) and basal PDMS composition.

I expected the thickness of MRE will influence magnetic field induced stiffness change. Consistent with my expectation, thickness is the most influential condition so far, we get a big elastic modulus change from 18kPa to 58kPa by changing the thickness of the MRE to 2.95mm, this induced change is good for future studies on cell behaviors on these substrates which has a big variation in stiffness. In control PDMS elastomer devices without any iron inclusions or magnetic field applied, I found that compression test results are generally higher than tensile test results reported in previous studies. However, unexpectedly, I found that even without magnetic field or inclusion of carbonyl particles, the Elastic modulus observed changed when the substrate thickness was decreased from 5.7mm to 2.95mm. This unexpected result requires further investigation.

Key words: Magnetorheological Elastomers (MRE), Polydimethylsiloxane (PDMS), Elastic Modulus
1. Introduction

Mechanical loading has a significant impact on cell behaviors such as proliferation and differentiation \cite{1,2}. Polyacrylamide (PA) hydrogels \cite{3} were used in seminal experiments to provide a 2D cell culture microenvironment of suitable elasticity. Substrates based on elastomers such as Polydimethylsiloxane (PDMS) may be more suitable in certain cases because of their longer shelf-life under ambient conditions, while PA hydrogels have a shorter shelf-life of only have a few days\cite{4}. In previous studies, especially for in studying diseases such as hypertrophic cardiomyopathy, physical properties such as stiffness and viscoelasticity of the Extracellular Matrix (ECM) are important considerations \cite{5}. Previously, for example, Raczkowska et al. \cite{6} used Sylgard\textsuperscript{TM}184 admixed with benzophenone, and they made a series of PDMS substrates of different stiffnesses. After culturing non-malignant (HCV29) and cancerous (T24) bladder cancer cells, they found that PDMS substrate elasticity has a significant contribution on the behavior of cells: softer PDMS substrate demonstrated excellent cytocompatibility whereas stiffer substrates repelled cells, where the proliferation rate decreased substantially.

![Figure 1 Composite consisting of the polymer matrix and iron particles. When a magnetic field is applied, the particles rearrange and cause a change in the material stiffness\cite{7}](Image)

However, both pure PDMS elastomers and PA hydrogels have a clear drawback: the stiffness of both systems is stable after creation, and this is not the case in real tissues that dynamically change their mechanical properties during development and disease\cite{7}. Therefore, developing a substrate in which stiffness can be externally controlled is a critical challenge. Corbin et al. \cite{8} introduced an approach of mixing magnetic carbonyl iron nanoparticles (CIPs) into PDMS elastomers (Figure 1). Using this approach, the stiffness of the substrates can be tuned by simply applying or removing magnets from the substrates. Experiments showed that the stiffness of the substrates can be increased and decreased by applying spacers to control the distance between the magnet and the substrate. Importantly, the Elastic Modulus (Young’s Module) of the resulting magnetized substrates only depends on the distance between the substrate and the magnet, and does not exhibit hysteresis (e.g. the modulus does not depend on whether the magnetic field intensity is going up or down). These magnetorheological elastomers (MREs) should be a good model of understanding the effects of mechanical load on how cells behave.

Here, my objective was to find a way to induce the largest possible magnet-induced change of the stiffness of the MREs. To achieve this goal, I adjusted several
conditions during the preparation of the MREs, including: 1) Thickness; 2) weight percent (wt %) of carbonyl iron particles (CIPs; 3) PDMS composition (ratio of part A and B in Sylgard™527, Dow Corning); 4) the storage time of the MREs. As a comparison to the MREs, pure PDMS substrates will also be made. According to previous reports, the stiffness of the PDMS substrates changes by mixing two different types of PDMS elastomers together with different ratios\textsuperscript{[9]} (Figure 2). Here, I replicated the PDMS substrates with the same conditions reported in the paper to make a comparison to the previous studies and the MREs on the stiffness. After substrate creation, indentation testing were performed on the samples.

![Figure 2](image)

**Figure 2** PDMS formulations span a wide range of mechanical properties from soft gels to stiff elastomers (Data in the graphs are all cited from the paper by Palchesko et al.\textsuperscript{[9]})

2. Experiment and Characterization

2.1 Creation of Magneto rheological Elastomers (MREs)

The base polymer of the MREs were made by Polydimethylsiloxane elastomers (Sylgard™527 part A and part B, product of Dow Corning, Ltd.), and Carbonyl Iron Powder (ChemicalStore, CAS: 7439-89-6) was added to the PDMS mixtures. Sylgard 527 parts A and B were mixed with a ratio of 1:1 in a weighing plate, followed by adding 50wt% of CIPs. The mixture was then stirred vigorously by hand for at least 10 minutes, then transferred into a 15mL centrifuge tube and sonicated at (xx parameters) for 3 minutes. Next, a PDMS mold ((20mm*20mm*5.7mm) was attached to an acrylic substrate with binder clips, and sprayed with Rain-X to prevent newly crosslinked PDMS from adhering to the frame or acrylic. Finally, the PDMS/CIP mixture was poured into the mold in a vacuum desiccator where it was subjected to vacuum for 10 minutes. Finally, the degassed PDMS/CIP mixture was baked in a 60\textdegree C oven for at least 24 hours.

In addition to the conditions mentioned above, I also made MREs under other conditions (thickness, wt% of the CIPs, ratio of different PDMS) was varied to investigate what kind of conditions can give us the biggest change in Elastic Modulus.

2.2 Creation of pure PDMS substrates

Pure PDMS substrates were created by combining two different PDMS elastomers (Sylgard™527 Parts A and B, Sylgard™184 and Sylgard™184 Curing Agent) with
different ratios. After adding different PDMS into the weighing plate, the mixtures were stirred for at least 10 minutes subjected to vacuum in a bell chamber, and finally baked at 60°C for at least 24 hours.

The ratios of the PDMS elastomers used are shown in Table 1:

<table>
<thead>
<tr>
<th>Sample #</th>
<th>Sylgard<strong>TM</strong>527</th>
<th>Sylgard<strong>TM</strong>184</th>
<th>Sylgard<strong>TM</strong>184 %</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>1</td>
<td>0</td>
<td>0%</td>
</tr>
<tr>
<td>2</td>
<td>40</td>
<td>1</td>
<td>2.439%</td>
</tr>
<tr>
<td>3</td>
<td>10</td>
<td>1</td>
<td>9.091%</td>
</tr>
<tr>
<td>4</td>
<td>5</td>
<td>1</td>
<td>16.667%</td>
</tr>
</tbody>
</table>

**Table 1** Composition of different PDMS products in samples. Each sample has a 1:1 ratio of part A and part B of Sylgard**TM**527, and a ratio 10:1 of Sylgard**TM**184 elastomer and Sylgard**TM**184 curing agent.

2.3 Indentation Testing on the substrates

Indentation testing was performed on the substrates using the Electroforce 3200 (TA instruments, Prof. Michelle Oyen of the MEMS department is appreciated for the supply and guidance of this device, Figure 3a). The radius of the spherical indent head is 6mm, and two types of load cells (45N and 2.5N) and magnets (N48 and N52) were used. Before performing the test, put the substrates on the cylindrical 3-D printed spacer containing the magnet and other plastic spacers, water repellent was added to the surface of the substrates to avoid the influence of adhesion between the indent head and the substrates, we can apply the magnetic force to the substrate by simply putting the magnet on the top of the spacer (Figure 3b).

![Figure 3 photos of the test device: a. photo of Electroforce 3200, where attachments (white cylinders) are added to the machine; b. photo of the spacers within the cylinder-shaped attachments, the black plastic spacers will separate the magnet from the substrates which sits on top the lid of the cylinder attachment on the bottom, when magnet force is needed, we can simply move the magnet to the top and put plastic spacers to the top.](image)

During the test, we first programmed the indentor head to go down and find the substrate, once contact was achieved, the head was immediately drawn back to zero force and allowed to dwell for ~30s before starting the actual test. The indent depth was chosen to not exceed 10% of the whole thickness of the substrate. The formula we used using to calculate the Elastic Modulus of the substrates is the Hertizan formula
shown in Figure 4. Within this formula, $F$ stands for the force received by the sample when it’s indented, $d$ stands for the displacement of the indenting process, $E^*$ stands for the Elastic Modulus and it meets the second equation in which $E_1$, $E_2$ are the moduli of the substrate and the indentor head. We consider $E_2$ to be infinite because it has an extremely-high modulus, and $v$ stands for the Poisson’s ratio, which we also consider this value to be constant 0.5, the Poisson’s ratio value of a perfect elastomer.

At least 3 MRE samples ($n=3$) of each condition were tested to ensure the robustness of the conclusions.

$$F = \frac{4}{3} E^* R^2 \frac{1}{d^2}$$  \hspace{1cm} (1)

$$\frac{1}{E^*} = \frac{1 - \nu_1^2}{E_1} + \frac{1 - \nu_2^2}{E_2}$$  \hspace{1cm} (2)

Figure 4 Formula used to calculate Elastic Modulus of the substrates, we consider the $E_2$ to be infinite and $\nu$ to be 0.5.

With this formula, I use a MATLAB code (created by David Schuftan from the Huebsch lab) to make calculations and output the displacement-time and load-time curves.

3. Results and Discussion
3.1 Elastic Modulus results of MREs with different conditions

3.1.1 Elastic Modulus results of MREs with different thickness

During the experiment phase, MREs with two different thickness, 5.7mm and 2.95mm, were created using the standard conditions (50wt% CIP, Sylgard 527A:B=1:1). Indentation testing results (Figure 5a) showed that 2.95mm substrates tends to have a larger magnet-induced change in the moduli $(18\pm0.81kPa\sim58\pm5.53kPa, n=3)$ comparing to 5.7mm ones $(9\pm1.30kPa\sim26\pm3.31kPa, n=3)$, this fits the initial hypothesis of our group, and the variation of nearly 3 times larger is acceptable and will be a good model for further studies on how cells behave to this huge change in the mechanical stiffness of the biomaterials it attached to.
3.1.2 Elastic Modulus results of MREs with different weight percentage of CIPs

MREs with 60wt% of carbonyl iron particles are created and done indentation test to make comparison to the results with the substrates containing 50wt% of CIP. Indentation testing results showed that, the elastic modulus of the substrates without magnet remains similar, showing that changing the wt% of CIP from 50 to 60% did not have a measurable impact on the baseline. However, once the substrates are magnetized, I observed a larger change in elastic modulus in both 5.7mm substrates (9±1.30~25±3.31kPa vs 11±1.67~39±2.57kPa, n=3) and 2.95 mm substrates (18±0.81kPa~58±5.53kPa vs 25±0.95~70±5.26kPa, n=3) (Figure 6), leading to the conclusion that the increase amount of carbonyl iron particles may lead to the increase in stiffness of the MREs.

3.1.3 Elastic Modulus results of MREs with magnetic force of different intensity

Indentation tests were performed on samples with a condition of 50wt% CIP, Sylgard 527 A:B=1:1, 2.95mm and 5.7mm with 2 different magnets: a 1/8-inch N48 with a magnet flux density around 1700Gs, and a 1/4-inch N52 around 3500Gs. It’s worth mentioning that we used the N48 magnet to test all the other results showed above, and our hypothesis is that we will get a bigger change in elastic modulus by using a magnet that is twice as strong than then previous one. However, it seems to be the case that the magnetic flux density did not affect the elastic modulus of the magnetized substrate no matter what thickness the substrate is, but changes did appear in the relaxation (appears to be a decrease in load on the graph) that has been seen with all tests with magnets. The relaxation is bigger with a
stronger magnet, and this relaxation is bigger on 5.7mm substrates than 2.95mm ones (Figure 7) However, we cannot conclude that the change of magnetic flux density does not have effect on the elastic modulus of the substrates, more tests such as repeating tests needs to be performed before we can confirm this conclusion (n=1 for now).

Figure 7 Elastic Modulus results of MREs (50wt% CIP, A:B=1:1, 5.7mm and 2.95mm) with different magnets: a. 5.7mm substrates magnetized with N48; b. 5.7mm substrates magnetized with N52; c. 2.95mm substrates magnetized with N48; d. 2.95mm substrates magnetized with N52; e. value of load decrease (relaxation) with two samples.

3.2 Elastic Modulus results of pure PDMS substrates

3.2.1 Elastic Modulus results of different ratios of PDMS

According to the experiments, we made 5.7mm pure PDMS substrates with different ratios of Sylgard 527 and Sylgard 184. After doing indentation tests on these samples, I plotted the results into a graph and added a tendency curve shown in Figure 8.

Figure 8 Tendency curve of Elastic modulus values of the PDMS substrates with different percentage of Sylgard 184 in the sample: a. tendency curve of indentation testing on substrates with different 184 %, the equation is \( y = 0.5753x^2 + 2.8291x + 7.7153 \) \( R^2 = 0.9984 \)
According to the tendency curve, we can see an increase in the elastic modulus of the pure PDMS substrates when we increase the ratio of Sylgard 184 in the mixture, this conclusion is consistent with the report of the Palchesko paper\cite{9}.

3.2.2 Research on the uniformity of Elastic modulus of pure PDMS substrates with different thickness

In order to confirm the uniformity of Elastic modulus of pure PDMS substrates with different thickness, PDMS substrates (184:527= 1:40, 2.95mm and 5.7mm thick) was made and crosslinked in the 60°C oven for over 24h. The substrates are performed with indentation tests with a indent depth of 10% of the whole substrate with the 45N load cell, surprisingly, the results showed that 2.95mm substrates has an elastic modulus of 36±5.63kPa, n=4, while the 5.7mm substrates is 12±2.34kPa, n=4 (Figure 9). It’s worth noting that according to Figure 2b, 15kPa should be the correct value for substrates with a 184 ratio of 2.43% (184:527=1:40), so the elastic modulus get with the thinner substrates is abnormal.

![Elastic modulus results of PDMS substrates with different thickness](image)

**Figure 9** Elastic modulus results of PDMS substrates with different thickness, conditions: 60°C baked, 45N load cell, indent depth 10%.

To further understand the trend, besides the two thicknesses we currently have, a super thick substrate (11.4mm) was made, and indentation tests of different indent depths from 0.3mm to 1.1mm with a step size of 0.2mm were performed on these super thick substrates. Similarly, indent depths from 0.1mm to 0.4mm with a step size of 0.1mm were performed on the 2.95mm substrates.
The results show that the 11.4mm sample remains at an elastic modulus of ~10kPa regardless of the indent depth, this value is close to the value get with the 5.7mm sample. However, the 2.95mm also get values close to 40kPa no matter how deep it is indented (Figure 10).

Figure 10 Elastic Modulus values of PDMS substrates (184:527=40:1) with a thickness of 11.4mm and 2.95mm: a. Modulus values of 11.4mm substrate indenting 0.3,0.5,0.7,0.9 and 1.1mm; b. Modulus values of 2.95mm substrate indenting 0.1,0.2,0.3 and 0.4mm.

As a solution to this problem of getting different modulus values on substrates with different thickness, a total of 3 different methods is tried: 1. Crosslink at a higher temperature; 2. Change indent depth; 3. Use different load cell when doing tests. To be specific, we tried to crosslink the states in the 110°C oven, change the indent depth during the tests to 0.3mm (same indent depth as the thinner substrate), and use a smaller load cell to eliminate the noise appeared in the tests to improve the accuracy of the value.

As shown in Figure 11, after three tries on the conditions, the unexpected variance in substrate elastic modulus with constant Sylgard composition but different thickness still remains. It seems that the thickness of the substrates is surely having some effects on the stiffness of the PDMS substrates, however, it requires further research since other kinds of tests needs to be performed before we can confirm.

Figure 11 Elastic Modulus Results after changing conditions: a. Modulus results of the 5.7mm substrates indenting in different depths; b. modulus results of the 5.7mm and 2.95mm substrates with different crosslink temperatures indented at a depth of 0.3mm.
4. Conclusion and Future studies

4.1 Conclusions

Based on the results got in the research, I report the following conclusions:

1) For the MREs, we managed to get a big elastic modulus change from 18±0.81kPa to 60±5.53kPa by making the substrate thinner (from 5.7mm to 2.95mm), this variation in Elastic Modulus is solid for the future studies on how cells behave under such big change in stiffness. Furthermore, I observed an increase in the change of elastic modulus by the increase of the wt% of the CIPs, where I see a change of 11±1.67~39±2.57kPa for 5.7mm substrates, and 25±0.95~70±5.26kPa for 2.95mm substrates.

2) For the pure PDMS substrates, an increase in the elastic modulus is seen when the ratio of Sylgard 184 is increased. The data fits a tendency curve of $y = 0.5753x^2 + 2.8291x + 7.7153$ ($R^2=0.9984$).

3) An important and unexpected trend I found was that there is a large effect of substrate thickness on the elastic modulus of substrates made with the same ratio of Sylgard 184 and Sylgard 527. This is an unexpected result since elastic modulus is an intrinsic material property. This variation remained the same across a range of different indentation depths, crosslink temperatures, or load cell used. However, this requires further research since a higher $n$ would be needed.

4.2 Future Studies

According to the conclusions and results, future studies of this project can be as follows:

1) Change other conditions (type of magnetic particles, high-level oxidation of the samples, ratio of PDMS elastomers, etc.) to see if a bigger variation in elastic modulus can be observed.

2) Perform the 2.95mm MRE made with different conditions in cell behavior studies (Ongoing).

3) Find an easy way to do shear testing on the substrates (Ongoing).

4) Find out the mystery of modulus change seen in pure PDMS when the substrates are thinner (Ongoing).
5. References

6. Appendix
Matlab code used for Elastic Modulus calculation (created by David Schuftan)

% Use this code to get the toughness, elastic modulus, max stress, and max
% strain from mechanical testing.
%
[files,pathname] = uigetfile('*.csv', 'Select One or More Files', 'MultiSelect', 'on');

if isa(files, 'char') == 1
    total = 1;
else
    total = max(size(files));
end

count = 1;

nfiles = cell(1,total);

if ispc
    delim = '\';
elseif ismac
    delim = '/';
end

if contains(pathname, '.is_comp_RawData')
    npathname = pathname(1:end-17);
    movefile(pathname,npathname)
    pathname = [npathname delim];
end

% thickCheck = 0;
% while thickCheck == 0
%     thick = str2double(cell2mat(inputdlg('Height of Gel (mm)?')));
%     if ~isempty(thick) && isa(thick,'double') && thick > 0
%         thickCheck = 1;
%     end
% end
% shape = questdlg('Specimen Cross-Section Shape?','Shape','Rectangle','Circle','Circle');
% switch shape
%     case 'Rectangle'
%         rectlength = str2double(cell2mat(inputdlg('Length of Sample (mm)?')));
%         rectlength = .001.*rectlength; % to meters
%         rectwidth = str2double(cell2mat(inputdlg('Width of Sample (mm)?')));
%         rectwidth = .001.*rectwidth; % to meters
%         area = rectlength*rectwidth; %m^2
%     case 'Circle'
%         diamCheck = 0;
%         while diamCheck == 0
%             diameter = str2double(cell2mat(inputdlg('Diameter of Sample (mm)?')));
%             if ~isempty(diameter) && isa(diameter,'double') && diameter > 0
%                 diamCheck = 1;
%             end
%         end
%         diameter = .001 * diameter;
%         area = pi.*(diameter/2)^2;
% end

indentradius = str2double(cell2mat(inputdlg('Radius of Indentor (mm)')));

while count <= total
    % choose specific file
    tic
    if isa(files, 'char') == 1
        filename = files;
    else
        filename = files{1,count};
    end
    disp(filename);
    nfilename = filename(1:end-8);
    filedata = readcell([pathname filename],'DatetimeType','text','LineEnding','\r');
    [r,c] = find(cellfun(@isnumeric,filedata) == 1);
    c = unique(c);
runique = unique(r);

for i = 1:length(runique)
    numrow = sum(r(:) == runique(i));
    if numrow ~= length(c)
        runique(i) = 0;
    end
end

runique(runique == 0 ) = [];
r = runique;

data1 = filedata((r(1):2:r(end),c(1):c(end));
data1(cellfun(@(x) isa(x,'missing'), data1)) = {nan};
gaps = cell2mat(cellfun(@isnan, data1(3:end,1), 'UniformOutput', false));
gaps = find(gaps);
gaps = gaps + 2;

header = string(data(1:2:r-1,c(1):c(end)));
data = cell2mat(data1(3:end,:));
correct0 = mean(data(gaps(2)-1:gaps(3)-2));
data_cor = data; data_cor(:,2) = data_cor(:,2) - correct0;

sec1 = data_cor(1:gaps(1)-3,:); % Initial indent to find surface
sec2 = data_cor(gaps(1)-1:gaps(2)-3,:); % Back up to top of surface
sec3 = data_cor(gaps(2)-1:gaps(3)-3,:); % Wait at top of surface for substrate to relax
sec4 = data_cor(gaps(3)-1:gaps(4)-3,:); % Indent surface prescribed amount
sec5 = data_cor(gaps(4)-1:end,:); % Wait at indented depth for any relaxation

E = 1000*((9/16).*abs(sec5(:,3))./((indentradius^(1/2)).*(abs(sec5(:,2)).^(3/2)))); %kPa

E_mean = mean(E);

fig = figure;
ax = axes(fig);
title(ax,nfilename(1:end-4),'Interpreter','none')
xlabel(ax,strcat(strtrim(header(1,1))," (",strtrim(header(2,1)),")"))
yyaxis(ax,'left')
plot(ax,fullTest_cor_stitch(:,1),fullTest_cor_stitch(:,2));
yyaxis(ax,'right')
plot(ax,fullTest_cor_stitch(:,1),fullTest_cor_stitch(:,3));
ylabel(ax,strcat(strtrim(header(1,3))," (",strtrim(header(2,3)),")"))
hold on
plot(ax,fullTest_cor_stitch(:,1),smooth(fullTest_cor_stitch(:,3),50),'--k');
yyaxis(ax,'left')
ylabel(ax,strcat(strtrim(header(1,2))," (",strtrim(header(2,2)),")"))
legend(ax,strcat(strtrim(strtrim(header(1,2))," (",strtrim(header(2,2)),")"),...
strcat(strtrim(header(1,3))," (",strtrim(header(2,3)),")"),...
strcat("Smoothed ",strtrim(header(1,3))," (",strtrim(header(2,3)),")"))
annotation(fig,'textbox',[
.1339 .119 .334 .056],'
Elastic Modulus:
',num2str(E_mean),'kPa'))

dirname = pathname;

if isequal(exist([dirname nfilename], 'dir'),0)
    mkdir([dirname nfilename])
else
    dirname = ([dirname nfilename delim]);
end

saveas(fig, [dirname nfilename '_Indent Fig.jpg'])
savefig(fig, [dirname nfilename '_Indent Fig.fig'])

if isequal(exist([pathname 'Indent Data'], 'dir'),0)
    mkdir([pathname 'Indent Data'])
else
    dirname = ([pathname 'Indent Data' delim]);
end
save([dirname nfilename '.mat'], 'E_mean')

count = count + 1;
end

%%% count2 = 1; addpath(dirname); tic
tic

files2 = dir(fullfile(dirname, '*.mat')); nfiles2 = {}; for i = 1:length(files2)
    nfiles2(i) = {files2(i).name};
end

total = length(nfiles2); splitpath = strsplit(pathname, delim);

% create empty arrays/vectors to be filled filenames = {}; % Added by Soore - empty cell array to be filled with filenames elasticModulus = zeros(total,1);

while count2 <= total
    filename2 = nfiles2{1,count2};

    load(fullfile(filename2)); disp(filename2)
    filenames(count2) = filename2(1:length(filename2)-4); elasticModulus(count2) = E_mean;

    count2 = count2 + 1;
end

filenames; elasticModulus;
% Create and Save Table into Folder where Data is Taken From

headers = {'filename','Elastic Modulus'};
units = {' ','kPa'};
nname = char(splitpath(length(splitpath)-1));
writecell(headers,[dirname name '.xlsx'],'Sheet',1,'Range','A1');
writecell(units,[dirname name '.xlsx'],'Sheet',1,'Range','A2');
writecell(filenames',[dirname name '.xlsx'],'Sheet',1,'Range','A3');
writematrix(elasticModulus,[dirname name '.xlsx'],'Sheet',1,'Range','B3');