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Supplementary Figure 1: Injection apparatus for microlymphangi-ography. After filling the entire apparatus with saline, a small air bubble can be introduced in the tip of the injection needle. Then the fluorescent dye can be drawn into the tubing, with the air bubble separating the fluorescent dye and the saline. The distance the air bubble travels and the cross sectional area of the tubing can be used to measure the volume of dye injected.

SUPPLEMENTAL INFORMATION

DIRECTIONS FOR RUNNING PEAK AND VALLEY ANALYSIS IN MATLAB

1. In Matlab, open new script.
2. In Matlab Editor, copy the script "PulsingTopFolder" from below and save as a MATLAB Code file "PulsingTopFolder.m".
3. Open new script and in Matlab Editor, copy the script "WorkOnXRange" from below and save as a MATLAB Code file "WorkOnXRange.m".
4. In Matlab Editor, copy the script "FinalVlymph" from below and save as a MATLAB Code file "FinalVlymph.m".
5. Upload saved .m files to Current Folder in Matlab
6. Create desktop folder entitled "working-on".
7. Place data file folder (containing series of acquired time-lapse images) into "working-on" folder
8. Within data file folder, create a new folder entitled "Matlab-results"
9. In the command window, type "PulsingTopFolder". Press enter.
10. Evaluate .avi videos to ensure that the entire length of the vessel segment is properly monitored for the duration of the video.
11. Create Excel document entitled "X_Range".
12. Save "X_Range" within the Matlab-results folder.
13. Indicate the horizontal region to be analyzed. For example, our images are 1344 pixels long. If the length of the vessel is correctly monitored for the entire video, indicate measurement should start at the first pixel and continue until pixel 1344.
14. In the command window, type "WorkOnXRange". Press enter.
15. In the command window, type "FinalVlymph". Press enter.
16. If peaks and valleys are properly identified,, type "1" at the prompt.
17. If peaks and valleys are not properly identified, type "0" at the prompt in order to manually set threshold for peaks and valleys.

SUPPLEMENTAL MATLAB CODE

PulsingTopFolder

```matlab
%% SECTION TITLE
% DESCRIPTIVE TEXT
close all;
clear all;

global start_x start_y img_size_x img_size_y gap_x gap_y
global bwareaopen_min
global avimovie source_folder


top_folder='../../../working-on/';

%%%%%%%%%%Input time step size%%%%%%%%%%
% % t_gap=0.08;
t_unit='sec';
%%%%%%%%%%Input time step size%%%%%%%%%%

%%%%%%%%%%Input time step size%%%%%%%%%%
img_size_x=336; %Image size in X
img_size_y=128; %Image size in X
%%%%%%%%%%Input time step size%%%%%%%%%%

gap_x=20;
gap_y=20;

start_x=100;
start_y=500;

bwareaopen_min=5000;

check_frame=0;


f = dir(top_folder); % you can specify the path also './path'
    % First two files grabbed on windows are '.' and '..'
    % therefore start with file 3
j=0;
for i=1:size(f,1)
    disp('------------------');
    tmp_name=f(i).name;
    tmp_length=length(tmp_name);

    if (tmp_length > 2)
%         if (tmp_name(tmp_length-2:tmp_length) == 'tif' & ...
%                 tmp_name(tmp_length-5:tmp_length-4) ~= '1X' & ...
%                 tmp_name(tmp_length-8:tmp_length-4) ~= 'stack')
%             j=j+1;
%             fnames{j}=tmp_name;
%         end
```

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```matlab
%              if (tmp_name(tmp_length-2:tmp_length) == 'tif')

        tmp_name
        if (length(strfind(tmp_name,'tmp')) == 0 & ...
            length(strfind(tmp_name,'Matlab')) == 0 & ...
            length(strfind(tmp_name,'.DS_Store')) == 0 & ...
            length(strfind(tmp_name,'._')) == 0)
%            if (tmp_name(tmp_length-5:tmp_length-4) ~= '1X')
%                if (tmp_name(tmp_length-8:tmp_length-4) ~= 'stack')
                  j=j+1;
                  sub_folders{j}=tmp_name;
%                end
%            end
        end

    end
end

clear f;

size(sub_folders,2);

for k1=1:size(sub_folders,2)

    f = dir(strcat(top_folder,sub_folders{k1})); % you can specify the path
also './path'
    % First two files grabbed on windows are '.' and '..'
    % therefore start with file 3
    j=0;
    for i=1:size(f,1)
        disp('--------------------');
        tmp_name=f(i).name;
        tmp_name
        tmp_length=length(tmp_name);

        if (tmp_length > 3)
%            if (tmp_name(tmp_length-2:tmp_length) == 'tif' & ...
%                    tmp_name(tmp_length-5:tmp_length-4) ~= '1X' & ...
%                    tmp_name(tmp_length-8:tmp_length-4) ~= 'stack')
%                j=j+1;
%                fnames{j}=tmp_name;
%            end

%                if (tmp_name(tmp_length-2:tmp_length) == 'tif')
            if (length(strfind(tmp_name,'tmp')) == 0 & ...
                length(strfind(tmp_name,'Matlab-results')) == 0 & ...
                length(strfind(tmp_name,'.DS_Store')) == 0 & ...
                length(strfind(tmp_name,'._')) == 0)
%                if (tmp_name(tmp_length-5:tmp_length-4) ~= '1X')
%                    if (tmp_name(tmp_length-8:tmp_length-4) ~= 'stack')
                      j=j+1;
```

```matlab
                    sub_sub_folders{j}=tmp_name;

%              end
%           end
        end

    end
end


result_folder=strcat(top_folder,sub_folders{k1},'/Matlab-results/');

    size(sub_sub_folders,2);


    for k2=1:size(sub_sub_folders,2)


        condition=sub_sub_folders{k2};

        tmp_length=length(condition);

        t_gap=str2num(condition(tmp_length-2:tmp_length))/1000;

XY_resolution=0.32225*20/str2num(condition(tmp_length-5:tmp_length-4))
;


        %%%%%%%%%Input where the image files are%%%%%%%%%

source_folder=strcat(top_folder,sub_folders{k1},'/',condition,'/');
        %%%%%%%%%Input where the image files are%%%%%%%%%

        %%%%%%%%%Input where the movie files should be saved%%%%%%%%%

        %%%%%%%%%Input where the movie files should be saved%%%%%%%%%


        % days=['Day 03';'Day 06';'Day 09';'Day 12';'Day 15';'Day 18';'Day
21';'Day 24'];
        %
        f = dir(source_folder); % you can specify the path also './path'
        % First two files grabbed on windows are '.' and '..'
        % therefore start with file 3
        j=0;
        for i=1:size(f,1)
            tmp_name=f(i).name;
            tmp_length=length(tmp_name);

            if (tmp_length > 8)
```

```matlab
%            if (tmp_name(tmp_length-2:tmp_length) == 'tif' & ...
%                  tmp_name(tmp_length-5:tmp_length-4) ~= '1X' & ...
%                  tmp_name(tmp_length-8:tmp_length-4) ~= 'stack')
%              j=j+1;
%              fnames{j}=tmp_name;
%            end


%            if (tmp_name(tmp_length-2:tmp_length) == 'tif')
            if (length(strfind(tmp_name,'-0')) ~= 0 & ...
               length(strfind(tmp_name,'._')) == 0)
%              if (tmp_name(tmp_length-5:tmp_length-4) ~= '1X')
%                  if (tmp_name(tmp_length-8:tmp_length-4) ~= 'stack')
                    j=j+1;
                    fnames{j}=tmp_name;
%                  end
%              end
            end

        end
    end

    if (j > 0)

        tot_steps=size(fnames,2);

        thrlds=0.1*ones(tot_steps,1);
        auto_dia=zeros(tot_steps,4);
%          manual_dia=zeros(tot_steps,4);
        prefix=fnames{1}(1:length(fnames{1})-5);

    % % % %    prefix=fnames{1}(1:length(fnames{1})-4);

        % show all files

        %%%%%%%%%Input file name for the current movie%%%%%%%%%
%
moviename=strcat(result_folder,condition,'@',prefix,'.avi');
%    fps=10;
%
avimovie=avifile(moviename,'QUALITY',100,'FPS',fps,'COMPRESSION','None');

        %%%%%%%%%Input file name for the current movie%%%%%%%%%
        main_fname=fnames{1}(1:length(fnames{1})-5);
        moviename=strcat(result_folder,condition,'.avi');

%
moviename=strcat(result_folder,condition,'@',prefix,'.avi');

        %%%%%%%%%Input file name for the current movie%%%%%%%%%
```

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```matlab
        fps=10;

avimovie=avifile(moviename,'QUALITY',100,'FPS',fps,'COMPRESSION','None
');

        for i=1:tot_steps

%          for i=1:10

            disp('#######################');
            i

% %          i=112

        % info=imfinfo('./Day09/0.5%W2P02-Day09-465um.tif');

            time=strcat([num2str((i-1)*t_gap),' ',t_unit]);

            if (i >=0 & i <= 9)
                step=strcat('000',num2str(i));
            elseif (i >=10 & i <= 99)
                step=strcat('00',num2str(i));
            elseif (i >=100 & i <= 999)
                step=strcat('0',num2str(i));
            elseif (i >=1000 & i <= 9999)
                step=strcat(num2str(i));
            else
                error('i out of range!');
            end


% % % %          if (i >=0 & i <= 9)
% % % %              step=strcat('00',num2str(i));
% % % %          elseif (i >=10 & i <= 99)
% % % %              step=strcat('0',num2str(i));
% % % %          elseif (i >=100 & i <= 999)
% % % %              step=strcat(num2str(i));
% % % %          else
% % % %              error('i out of range!');
% % % %          end


        %%%%%%%%%Input the main file name for the images%%%%%%%%%
        IMG_file=strcat(source_folder,fnames{i});
        %%%%%%%%%Input the main file name for the images%%%%%%%%%

        IMG=imread(IMG_file);
```

```matlab
                IMG_min=min(min(double(IMG)));
                IMG_max=max(max(double(IMG)));

                IMG=(double(IMG)-IMG_min)/(IMG_max-IMG_min);

                [npx npy]=size(IMG);


[auto_results,new_thrld]=Pulsing_Threshold_Vessel_New_2img_NoManual(ch
eck_frame,i,IMG,thrlds(i));

                if (i == 1)
                    auto_all_frames=uint8(zeros(npx,npy,tot_steps));
%                    manual_all_frames=uint8(zeros(npx,npy,tot_steps));
                end


                auto_all_frames(:,:,i)=auto_results;
%                  manual_all_frames(:,:,i)=manual_results;


        %          avimovie=addframe(avimovie,frame);

                thrlds(i)=new_thrld;

                [auto_area,auto_angle]=Get_Area_Angle(auto_results);
                auto_dia(i,1)=i;
                auto_dia(i,2)=(i-1)*t_gap;

                if (auto_area > 0)

auto_dia(i,3)=auto_area/(npy/cosd(auto_angle))*XY_resolution;
                else
                    auto_dia(i,3)=0;
                end

%
[manual_area,manual_angle]=Get_Area_Angle(manual_results);
%                manual_dia(i,1)=i;
%                manual_dia(i,2)=(i-1)*t_gap;
%
%                if (manual_area > 0)
%
manual_dia(i,3)=manual_area/(npy/cosd(manual_angle))*XY_resolution;
%                else
%                    manual_dia(i,3)=0;
%                end

            end
```

```matlab
        disp('&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&');
        disp('&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&&');
        disp('Making the auto_all_frames movie. Please wait ...');

        AllFrames_Movie(result_folder,condition,auto_all_frames);


%     avimovie=close(avimovie);

        % % % % % % % % % % % % % % % % % % % % % % % % % % % % % % % % %
        auto_dia(:,4)=auto_dia(:,3)/auto_dia(1,3);
        figure;
        plot(auto_dia(:,2),auto_dia(:,3),'-');
        ylim([0 200])
        title([condition,'@',prefix,',
auto'],'FontSize',16,'FontWeight','bold','Color','blue');
        xlabel(['Time, s'],'FontSize',14,'FontWeight','bold');
        ylabel(['Vessel Diameter,
um'],'FontSize',14,'FontWeight','bold');
        set(gca,'FontSize',12,'FontWeight','bold');
        set(gcf,'Color',[1 1 1]);

        matlab_tif=strcat(result_folder,condition,'-auto.tif');
        set(gcf,'PaperPositionMode','auto');   % Use screen size
        print('-dtiffn',matlab_tif);
        close;

        results_fname=strcat(result_folder,condition,'-auto.txt');
        save(results_fname,'auto_dia','-ASCII');

        % % % % % % % % % % % % % % % % % % % % % % % % % % % % % % % % %
%        manual_dia(:,4)=manual_dia(:,3)/manual_dia(1,3);
%        figure;
%        plot(manual_dia(:,2),manual_dia(:,3),'-');
%        ylim([0 200])
%        title([condition,'@',prefix,',
manual'],'FontSize',16,'FontWeight','bold','Color','blue');
%        xlabel(['Time, s'],'FontSize',14,'FontWeight','bold');
%        ylabel(['Vessel Diameter,
um'],'FontSize',14,'FontWeight','bold');
%        set(gca,'FontSize',12,'FontWeight','bold');
%        set(gcf,'Color',[1 1 1]);
%
%
matlab_tif=strcat(result_folder,condition,'@',prefix,'-manual.tif');
%        set(gcf,'PaperPositionMode','auto');   % Use screen size
%        print('-dtiffn',matlab_tif);
%        close;
%
%
results_fname=strcat(result_folder,condition,'@',prefix,'-manual.txt')
```

```matlab
;
%               save(results_fname,'manual_dia','-ASCII');
%
%
thrlds_fname=strcat(result_folder,condition,'@',prefix,'-manual-thrlds
.txt');
%               save(thrlds_fname,'thrlds','-ASCII');



all_frames_fname=strcat(result_folder,condition,'-AllFrames.mat');
            save(all_frames_fname,'auto_all_frames');


        %
manual_all_frames_fname=strcat(result_folder,condition,'@',prefix,'-Ma
nualAllFrames.mat');
        %       save(manual_all_frames_fname, 'manual_all_frames')

            avimovie=close(avimovie);

            clear fnames;

        end

    end

    clear sub_sub_folders;

end
```

B6$_M$8$_0$1$_2$0X080@B6$_M$8$_0$1$_2$0X080-0001, auto
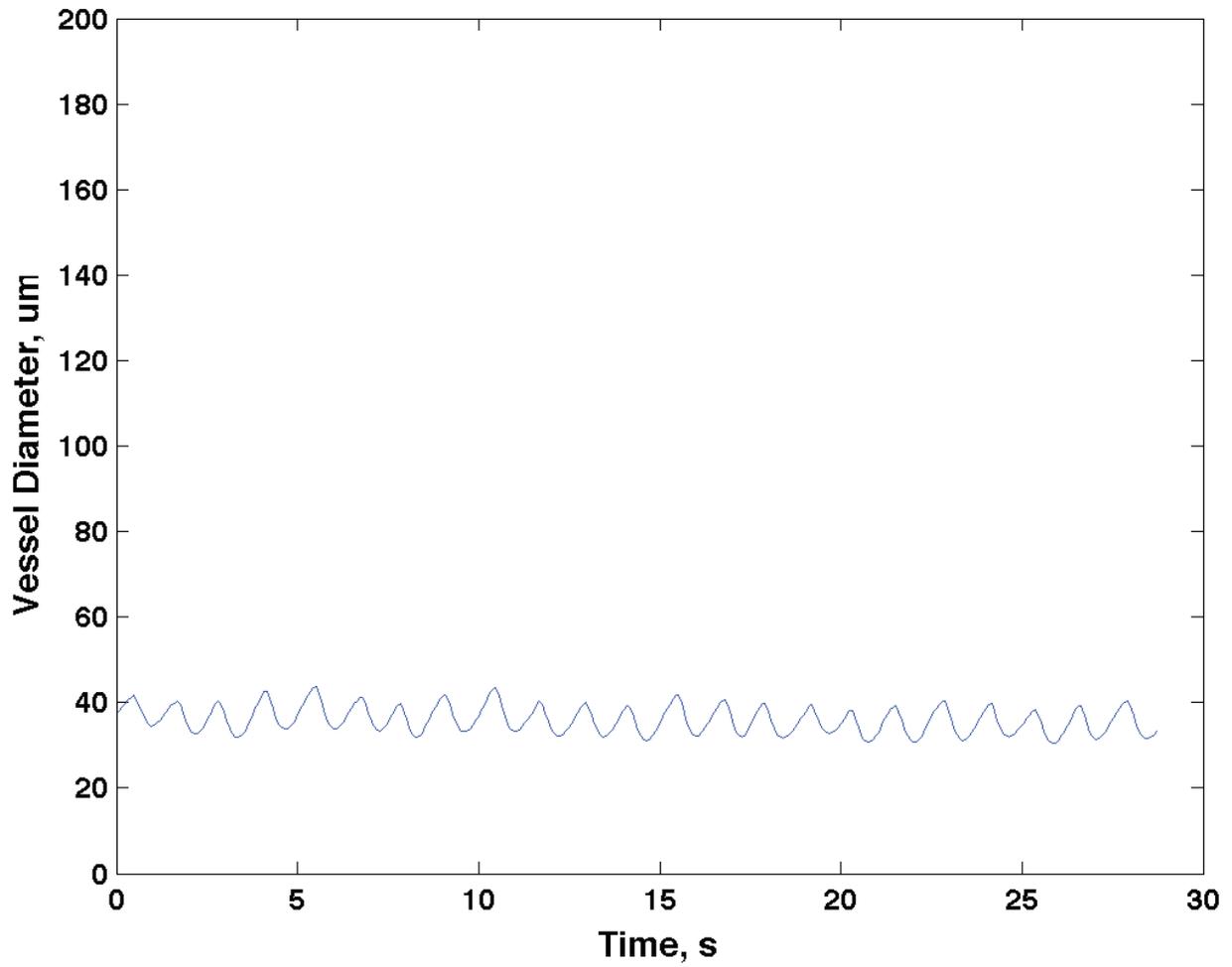
SUPPLEMENTAL MATLAB CODE

WorkOnXRange

```matlab
clear all;
close all;

pi=3.14159265;

img_size_x=512; %Image size in X
img_size_y=256; %Image size in X
%%%%%%%%%Input time step size%%%%%%%%%

gap_x=50;
gap_y=50;



start_x=100;
start_y=500;

top_folder='../../../working-on/';

% sub_folders=struct('name',{'061708B6M3_5','061008C57B6M1_2'});

f = dir(top_folder); % you can specify the path also './path'
    % First two files grabbed on windows are '.' and '..'
    % therefore start with file 3
j=0;
for i=1:size(f,1)
    disp('-------------------');
    tmp_name=f(i).name;
    tmp_length=length(tmp_name);

    if (tmp_length > 2)
%         if (tmp_name(tmp_length-2:tmp_length) == 'tif' & ...
%                 tmp_name(tmp_length-5:tmp_length-4) ~= '1X' & ...
%                 tmp_name(tmp_length-8:tmp_length-4) ~= 'stack')
%             j=j+1;
%             fnames{j}=tmp_name;
%         end

%             if (tmp_name(tmp_length-2:tmp_length) == 'tif')

        tmp_name
        if (length(strfind(tmp_name,'tmp')) == 0 & ...
            length(strfind(tmp_name,'Matlab')) == 0 & ...
            length(strfind(tmp_name,'.DS_Store')) == 0 & ...
            length(strfind(tmp_name,'._')) == 0)
%             if (tmp_name(tmp_length-5:tmp_length-4) ~= '1X')
%                 if (tmp_name(tmp_length-8:tmp_length-4) ~= 'stack')
                j=j+1;
                sub_folders{j}=tmp_name;
%             end
%         end
```

```matlab
        end

    end
end

clear f;


for k=1:size(sub_folders,2)

% for k=1:1

    %%%%%%%%%Input where the image files are%%%%%%%%%
    folder=strcat(top_folder,sub_folders{k},'/Matlab-results/');


    X_Range_fname=strcat(folder,'X_Range.xls');
    [xrange,foldername,raw] = xlsread(X_Range_fname,'X_Range');
    foldername=char(foldername);

%    figure(1);
%    rect=[start_x start_y 2*img_size_x+4.5*gap_x img_size_y+2.5*gap_y];
%    set(gcf,'Units','pixels','Position',rect);

% % %     avg_FreqAmp=zeros(size(xrange,1),4);

    for i=1:size(xrange,1)

        trimmed_foldername=strtrim(foldername(i,:));

%        foldername(i,:)

        tmp_length=length(trimmed_foldername)
        t_gap=str2num(trimmed_foldername(tmp_length-2:tmp_length))/1000

XY_resolution=0.32225*20/str2num(trimmed_foldername(tmp_length-5:tmp_l
ength-4))

%        pause;

        x_start=xrange(i,1);
        x_end=xrange(i,2);

% % %         target_foldername=trimmed_foldername;
% % %         target_str='-AllFrames-FreqAmpValPeak.mat';
% % %         tmp_fname=Find_File(folder,target_foldername,target_str)
% % %
% % % %          pause;
% % %
% % %         load(tmp_fname);
```

```matlab
% % %
% % %          avg_FreqAmp(i,1)=x_start;
% % %          avg_FreqAmp(i,2)=x_end;
% % %
avg_FreqAmp(i,3)=mean(avg_frequency_amplitude(x_start:x_end,1));
% % %
avg_FreqAmp(i,4)=mean(avg_frequency_amplitude(x_start:x_end,2));

        target_foldername=trimmed_foldername;
        target_str='-AllFrames.mat';
%         tmp_fname=Find_File(folder,target_foldername,target_str);

        tmp_fname=strcat(folder,target_foldername,target_str);

%          pause;

        load(tmp_fname);

        dia_volume_T=zeros(size(auto_all_frames,3),3);

        for time=1:size(auto_all_frames,3)

            time

            dia_volume_T(time,1)=(time-1)*t_gap;


[auto_area,auto_angle]=Get_Area_Angle(auto_all_frames(:,x_start:x_end,
time));

            if (auto_area > 0)

dia_volume_T(time,2)=auto_area/((x_end-x_start)/cosd(auto_angle))*XY_r
esolution;
            else
                error('Chosen part of vessel has auto_dia=0!');
            end



dia_volume_T(time,3)=(x_end-x_start)/cosd(auto_angle)*XY_resolution*pi
*(dia_volume_T(time,2)/2.0)^2;

        end

        figure;

        [AX,H1,H2] =
plotyy(dia_volume_T(:,1),dia_volume_T(:,2),dia_volume_T(:,1),dia_volum
e_T(:,3),'plot');
```

```matlab
        set(get(AX(1),'Ylabel'),'String','Vessel Average Diameter,
um','FontSize',16,'FontWeight','bold');
        % set(get(AX(1),'xlim'),[0 150]);

        set(get(AX(2),'Ylabel'),'String','Vessel Volume,
um^3','FontSize',16,'FontWeight','bold');

        set(AX(1),'YColor','r','FontSize',12,'FontWeight','bold');
        set(AX(2),'YColor','b','FontSize',12,'FontWeight','bold');

        %
set(AX(2),'YColor','b','FontSize',12,'FontWeight','bold','XLim',[0
30],'XTick',[0:10:30]);

        xlabel('Time, sec','FontSize',16,'FontWeight','bold');

%        title([sphd_fnames{i},', no
control'],'FontSize',20,'FontWeight','bold');


set(H1,'LineStyle','-','Color','r','LineWidth',0.5,'Marker','s','Marke
rFaceColor','r','MarkerSize',3);

set(H2,'LineStyle','--','Color','b','LineWidth',0.5,'Marker','o','Mark
erFaceColor','b','MarkerSize',3);

% % %        plot(dia_volume_T(:,1),dia_volume_T(:,2),'--*');
% % %        xlabel('Time, sec','FontSize',12,'FontWeight','bold');
% % %        ylabel('Vessel Volume,
% um^3','FontSize',12,'FontWeight','bold');
% % %        title('Temporal Change of Vessel Volume in Chosen
Segment','FontSize',14,'FontWeight','bold','Color','blue');


matlab_tif=strcat(folder,trimmed_foldername,'-DiaVol_VS_Time4ChosenX.t
if');
        set(gcf,'PaperPositionMode','auto');   % Use screen size
        print('-dtiffn',matlab_tif);
        close;


tmp_fname=strcat(folder,trimmed_foldername,'-DiaVol_VS_Time4ChosenX.tx
t');
        save(tmp_fname,'dia_volume_T','-ASCII');

    end

% %    tmp_fname=strcat(folder,'All_Avg_FreqAmp_From_Dia.txt');
% %    save(tmp_fname,'avg_FreqAmp','-ASCII');
```
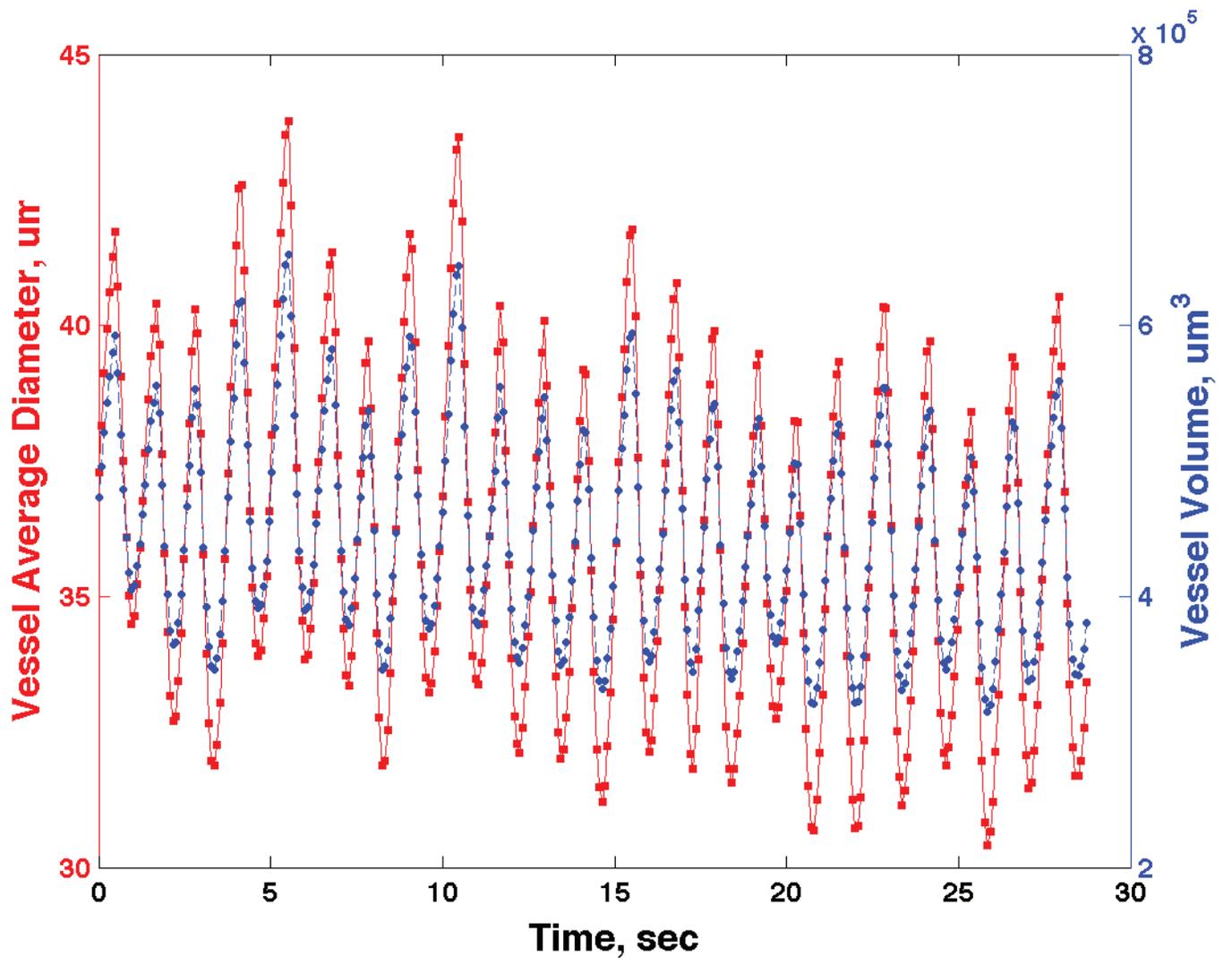
```matlab
% % % %      fname_tmp=strcat(folder,'All_Avg_FreqAmp_From_Dia.txt');
% % % %      fid = fopen(fname_tmp,'w');
% % % %      fprintf(fid,'%s \n','x_start  x_end      Avg Freq       Avg
Amp');
% % % %      for i=1:size(avg_FreqAmp,1)
% % % %
fprintf(fid,'%4.0f   %7.0f    %12.6f    %12.6f\n',avg_FreqAmp(i,:));
% % % %      end
% % % %      fclose(fid);


%     close(figure(1));

    clear fnames;

end
```

SUPPLEMENTAL MATLAB CODE

FinalVlymph

```matlab
clear all;
close all;

for x = 1:10
    disp(x)
end

pi=3.14159265;

img_size_x=512; %Image size in X
img_size_y=256; %Image size in X
%%%%%%%%%%%Input time step size%%%%%%%%%%

gap_x=50;
gap_y=50;


start_x=100;
start_y=500;

top_folder='../../../working-on/';

% sub_folders=struct('name',{'061708B6M3_5','061008C57B6M1_2'});

f = dir(top_folder); % you can specify the path also './path'
    % First two files grabbed on windows are '.' and '..'
    % therefore start with file 3
j=0;
for i=1:size(f,1)
    disp('-------------------');
    tmp_name=f(i).name;
    tmp_length=length(tmp_name);

    if (tmp_length > 2)
%         if (tmp_name(tmp_length-2:tmp_length) == 'tif' & ...
%                 tmp_name(tmp_length-5:tmp_length-4) ~= '1X' & ...
%                 tmp_name(tmp_length-8:tmp_length-4) ~= 'stack')
%             j=j+1;
%             fnames{j}=tmp_name;
%         end

%             if (tmp_name(tmp_length-2:tmp_length) == 'tif')

        tmp_name
        if (length(strfind(tmp_name,'tmp')) == 0 & ...
            length(strfind(tmp_name,'Matlab')) == 0 & ...
            length(strfind(tmp_name,'.DS_Store')) == 0 & ...
            length(strfind(tmp_name,'._')) == 0)
%             if (tmp_name(tmp_length-5:tmp_length-4) ~= '1X')
%                 if (tmp_name(tmp_length-8:tmp_length-4) ~= 'stack')
                j=j+1;
                sub_folders{j}=tmp_name;
%                 end
```

```matlab
%           end
        end

    end
end

clear f;

for k=1:size(sub_folders,2)

    %%%%%%%%%%Input where the image files are%%%%%%%%%%
    folder=strcat(top_folder,sub_folders{k},'/Matlab-results/');

    X_Range_fname=strcat(folder,'X_Range.xls');
    [xrange,foldername,raw] = xlsread(X_Range_fname,'X_Range');
    foldername=char(foldername);

% %      Dia_thrld, Avg_Dia_min, Avg_Dia_max, Avg_Freq, Avg_Amp, V_in/t, V_out/t

    final_results=zeros(size(xrange,1),9);

    figure(1);
    rect=[start_x start_y img_size_x+2*gap_x img_size_y+2*gap_y];
    set(gcf,'Units','pixels','Position',rect);

    for i=1:size(xrange,1)

        disp('#######################');

        trimmed_foldername=strtrim(foldername(i,:))

        tmp_length=length(trimmed_foldername)
        t_gap=str2num(trimmed_foldername(tmp_length-2:tmp_length))/1000
        XY_resolution=0.32225*20/str2num(trimmed_foldername(tmp_length-5:tmp_length-4))

% % %          fname=strcat(folder,trimmed_foldername,'-DiaVol_VS_Time4ChosenX.txt');

        target_foldername=trimmed_foldername;
        target_str='-DiaVol_VS_Time4ChosenX.txt';
        fname=Find_File(folder,target_foldername,target_str)

        y=load(fname);
        Dia=y(:,2);
        Vol=y(:,3);
        Length=Vol./(pi*(Dia./2).^2);

        mean_vessel_size=mean(Dia);

        peakdet_thrld_Dia=1;

        flag=0;

        while (flag == 0)
```

```matlab
        figure(1);

        rect_axes(1:4)=[gap_x gap_y img_size_x img_size_y];
        set(gca,'Units','pixels','Position',rect_axes);

        plot(Dia,'b-');

        [maxtab, mintab] = peakdet(Dia,peakdet_thrld_Dia);

        if (size(mintab,2) == 2 & size(maxtab,2) == 2)
            hold on; plot(mintab(:,1), mintab(:,2), 'g*');
            plot(maxtab(:,1), maxtab(:,2), 'r*');

            title(['current peakdet-thrld-Dia = ',num2str(peakdet_thrld_Dia)],'FontSize',16,'FontWeight','bold','Color','blue');
            xlabel('Time, sec','FontSize',14,'FontWeight','bold');
            ylabel('Vessel Diameter, um','FontSize',14,'FontWeight','bold');

            figure(1);
        else
            disp('peakdet_thrld_Dia too big, no valley or peak detected!');
        end

        flag=input('peakdet results good? 0-NO, 1-YES: ');

        if (flag == 0)

            peakdet_thrld_Dia=input('New peakdet-thrld-Dia: ');
            clf;
        end
    end

    matlab_tif=strcat(strcat(folder,trimmed_foldername,'-Chosen-Peakdet_thrld-Dia.tif'));
    set(gcf,'PaperPositionMode','auto');    % Use screen size
    print('-dtiffn',matlab_tif);

    clf;

% % % % Diameter threshold
    final_results(i,1)=peakdet_thrld_Dia;

% % % % Average minimum diameter
    final_results(i,2)=mean(mintab(:,2));

% % % % Average maximum diameter
    final_results(i,3)=mean(maxtab(:,2));

% % % % Frequency (calculated with just valleys)
    final_results(i,4)=mean(mintab(2:length(mintab),1)-mintab(1:length(mintab)-1,1));
    final_results(i,4)=1/(final_results(i,4)*t_gap);
```

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```matlab
% % % % Amplification
        final_results(i,5)=((mean(abs(maxtab(:,2)-mean_vessel_size))+ ...
            mean(abs(mintab(:,2)-mean_vessel_size)))/2)/mean_vessel_size;

% % % % V_in/t and V_out/t (volume of lymph per volume of vessel per unit time)

%           [Vin_vs_V_vs_T,Vout_vs_V_vs_T]=Integrate_Volume(y(:,1),Vol,t_gap);

        [Vin_vs_V_vs_T,Vout_vs_V_vs_T,Vin_vs_L_vs_T,Vout_vs_L_vs_T]=Integrate_Volume(y(:,1),Vol,Length,t_gap);


        final_results(i,6)=Vin_vs_V_vs_T;
        final_results(i,7)=Vout_vs_V_vs_T;

        final_results(i,8)=Vin_vs_L_vs_T;
        final_results(i,9)=Vout_vs_L_vs_T;

    end

    fname_tmp=strcat(folder,'All_FinalResults.txt');
    fid = fopen(fname_tmp,'w');
    fprintf(fid,'%s \n','Dia_thrld   Avg_Dia_min    Avg_Dia_max    Avg_Freq       Avg_Amp      V_in/V/t        V_out/V/t    V_in/L/t       V_out/L/t'
    for i=1:size(final_results,1)
        fprintf(fid,'%8.4f  %12.6f  %12.6f  %12.6f  %12.6f  %12.6f  %12.6f  %12.6f  %12.6f\n',final_results(i,:));
    end
    fclose(fid);

    close(figure(1));

    clear fnames;

    disp(['Folder: ',strcat(top_folder,sub_folders{k}),' finished! Continuing to the next one ...']);

end
```

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current peakdet-thrld-Dia = 1