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# Characterization of Tissue Sample- Muller Matrix Imaging

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Abstract: Polarimetric Imaging is an emerging technique that uses polarized light to probe the physical and physiological properties of tissues. Polarization Ellipsometric techniques are utilized to understand the variation of polarization properties of light through sample tissue. The obtained optical signature matrix helps clinicians understand and assess the tissue properties and provide knowledge about the functioning of the tissue. In our work tissues collected from pathology lab of Gandhi Medical College, Hyderabad, India is used for recognizing the signature matrix of the tissue. Conditions to obtain a recognizable Mueller Matrix with rotating optics are addressed and are used to understand the polarization variation in the tissue through a novel pixel by pixel analysis technique and via the normalized signature matrix of the tissue.

Key Words: - Mueller Matrix imaging, Optical signature, Polarization and Tissue imaging.

### 1. INTRODUCTION

Widely used technique of polarization measurement in which a linear variation between the polarization state of incident and excited beams from a sample are obtained is Mueller polarimetry; it describes the polarization properties of the sample by using a Polarization State Generator (PSG) and Polarization State Analyzer (PSA) with rotating wave plates, polarizer and analyzer. The polarization effects that take place in the sample of interest are understood in terms of the images obtained with predefined polarization states.

These images are further processed to understand the characteristic change in the image and are attributed to the variation in composition of the sample.

Mueller matrix measurement system was described in the works of some researchers [1-6]. The optical polarization effects simultaneously occurring in the sample are studied with respect to the 49 captured images and then reduced to 16 element Mueller matrix images. These elements obtained are processed by MatLab codes for processing images pixel by pixel and are then normalized to first element to get a signature matrix.

### 2. THEORY

The polarization state of light is characterized by stokes parameters represented [9] as



A light beam represented by this Stokes vector when incident on an optical element (here the tissue sample), undergoes a transformation represented by a 4 X 4 matrix called the Mueller or Polarization matrix and the transformed Stokes vector is written [9] as

$$S' = M \times S \qquad (2)$$

$$\begin{pmatrix} S'_{0} \\ S'_{1} \\ S'_{2} \\ S'_{3} \end{pmatrix} = \begin{pmatrix} m_{11} & m_{12} & m_{13} & m_{14} \\ m_{21} & m_{22} & m_{23} & m_{24} \\ m_{31} & m_{32} & m_{33} & m_{34} \\ m_{41} & m_{42} & m_{43} & m_{44} \end{pmatrix} \begin{pmatrix} S_{0} \\ S_{1} \\ S_{2} \\ S_{3} \end{pmatrix} = (3)$$
Where
$$\begin{pmatrix} S'_{0} \\ S'_{1} \\ S'_{2} \\ S'_{3} \end{pmatrix}$$
is the stoke vector of

transmitted/reflected/scattered light.

$$\begin{pmatrix} m_{11} & m_{12} & m_{13} & m_{14} \\ m_{21} & m_{22} & m_{23} & m_{24} \\ m_{31} & m_{32} & m_{33} & m_{34} \\ m_{41} & m_{42} & m_{43} & m_{44} \end{pmatrix}$$
 is the Mueller matrix and 
$$\begin{pmatrix} S_0 \\ S_1 \\ S_2 \\ S_3 \end{pmatrix}$$
 is the stoke vector of incident light.

This S' matrix gives measurable polarization information of the output light beam. The Mueller matrix M can be also written [7, 8] as



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Where

$$= \frac{1}{m_{11}} (m_{12} \quad m_{13} \quad m_{14})^T \&$$
$$\overline{P} = \frac{1}{m_{11}} (m_{21} \quad m_{31} \quad m_{41})$$

 $\overline{D}^T$ 

D =

are called as Diattenuation and Polarizance vector respectively and 'm' is a 3 X 3 matrix [4].

Diattenuation characterizes the intensity transmittances of the incident polarization states. The diattenuation takes values from 0 to 1 and is defined as,

$$D = \frac{T_{\max} - T_{\min}}{T_{\max} + T_{\min}}$$
(5)

All Mueller matrices may not be physically realizable. The basic constraint for a Mueller matrix<sup>10</sup> to be physically realizable is that.

$$(MM^{T})^{T} = \sum_{i,j=0}^{3} m_{ij}^{2} \le 4m_{00}^{2}$$
 ------(6)

This imposes a condition that the Degree of Polarization is less than or equal to one, i.e.,

$$P = \frac{\sqrt{\left(S1^2 + S2^2 + S3^2\right)}}{S0} \le 1 \quad \dots \quad (7)$$

The equal sign applies for non-depolarizing systems and the inequality otherwise. In our earlier communications [14] we have proved that we get a recognizable Mueller matrix for these wood samples, here now we are doing it for the breast tissue.

Retardance is a property that indicates the phase change and has constant intensity transmittance for any incident polarization state and is described by

$$\bar{R} \equiv R\hat{R} = \begin{pmatrix} Ra_1 \\ Ra_2 \\ Ra_3 \end{pmatrix} \equiv \begin{pmatrix} R_H \\ R_{45} \\ R_C \end{pmatrix} - \dots (8)$$

Where, the components indicate, horizontal,  $45^{\circ}$  and circular retardance respectively. The net linear retardance is

$$R_L = \sqrt{R_H^2 + R_{45}^2} \quad \dots \qquad (9)$$

and the total retardance is

$$R = \sqrt{R_{H}^{2} + R_{45}^{2} + R_{C}^{2}} = \sqrt{R_{L}^{2} + R_{C}^{2}} = \left|\vec{R}\right| - \cdots - (10)$$
  
3. EXPERIMENTAL PROCEDURE

The normal breast tissue sample was illuminated with laser of 20mW power and 632.8nm wavelength with predefined polarization state. The collection optics is kept at  $45^{\circ}$  from the input beam direction throughout the experiment. As shown in Figure 1.



The samples considered here are pathology tissues. The specimens selected for breast tissue processing is sent in 10% formalin for proper fixation of the tissue. Unfixed and autolysed tissues cannot be processed. The fixed specimen is grossed accordingly and specific sites for breast tissue processing have to be identified. Then using a scalpel blade the concerned part of the specimen is cut into even square shaped bits and taken in to capsules. These capsules are then taken through different concentrations of alcohol for dehydration and hardening of the breast tissue. It is inserted into containers with molten wax for impregnation of the wax into the tissues. This is to harden the tissue to prepare blocks which are later cut. The impregnated breast tissues are then blocked with molten wax so that the cut surface of the breast tissue is properly aligned and then allowed to harden. These blocks are mounted onto a micro tome which cuts the breast tissue in the blocks in to 3-4 microns thick sections [11, 12]. These sections are taken on to slides and stained with Hematoxylin and Eosin stains and is as shown in Figure2.



Fig.2. Breast Tissue sample used for experiment

A beam of photons from the source through a PSG was made to fall on the sample material and reflected beam was made to pass through PSA and falls on the CCD detector connected to a computer that records the intensity information. By varying the optical elements in the PSG and PSA, 49 different intensity images were recorded [10].

49 intensity images with various orientations of Polarizer and Analyzer are necessary to obtain the 16 elements of Mueller matrix images [13]. The 49 intensity images obtained are named as follows

I <sub>OO</sub>	I <sub>OH</sub>	I <sub>OV</sub>	I <sub>OP</sub>	I <sub>OM</sub>	I <sub>OR</sub>	I <sub>OL</sub>
$I_{\text{HO}}$	$\mathbf{I}_{\mathrm{HH}}$	$I_{\rm HV}$	$\mathbf{I}_{\mathrm{HP}}$	$\mathbf{I}_{\mathrm{HM}}$	$I_{HR}$	$\mathbf{I}_{\mathrm{HL}}$
$I_{VO}$	$I_{VH} \\$	$I_{VV}$	$I_{VP}$	$I_{VM}$	$I_{\text{VR}}$	$I_{VL}$
$I_{PO}$	$\mathbf{I}_{\mathrm{PH}}$	$I_{\rm PV}$	$\mathbf{I}_{\mathrm{PP}}$	$I_{\text{PM}}$	$I_{PR}$	$I_{\text{PL}}$
$I_{MO}$	$I_{\rm MH}$	$I_{MV}$	$I_{MP}$	$I_{MM}$	I <sub>MR</sub>	$I_{ML}$



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 $I_{RO} \quad I_{RH} \quad I_{RV} \quad I_{RP} \quad I_{RM} \quad I_{RR} \quad I_{RL}$ 

 $I_{LO} \quad I_{LH} \quad I_{LV} \quad I_{LP} \quad I_{LM} \quad I_{LR} \quad I_{LL}$  where, the first subscript indicates input state(PSG), and the second subscript the output state (PSA).

After acquiring 49 intensity images, the 16 elemental Mueller matrix images are obtained by using the expressions which are as given below.

 $\begin{array}{c} m_{11} = I_{OO}; \ m_{12} = I_{HO} - I_{VO} \\ m_{13} = I_{PO} - I_{MO}; \ m_{14} = I_{LO} - I_{RO} \\ m_{21} = I_{OH} - I_{OV}; \ m_{22} = (I_{HH} + I_{VV}) - (I_{HV} + I_{VH}) \\ m_{23} = (I_{PH} + I_{MV}) \ ) - (I_{PV} + I_{MH}); \ m_{24} = (I_{RV} + I_{LH}) - (I_{RH} + I_{LV}) \\ m_{31} = I_{OP} - I_{OM} \\ m_{32} = (I_{HP} + I_{VM}) - (I_{HM} + I_{VP}); \ m_{33} = (I_{PP} + I_{MM}) - (I_{PM} + I_{MP}) \\ m_{34} = (I_{RM} + I_{LP}) - (I_{RP} + I_{LM}); \ m_{41} = I_{OL} - I_{OR} \\ m_{42} = (I_{HL} + I_{VR}) - (I_{HR} + I_{VL}); \ m_{43} = (I_{PL} + I_{MR}) - (I_{PR} + I_{ML}) \\ m_{44} = (I_{RR} + I_{LL}) - (I_{RL} + I_{LR}) \end{array}$ 

#### 4. RESULTS

The 49 intensity images captured are used to obtain the 16 elemental Mueller matrices using the expressions above, these images are as shown in Figure 3.





Fig.3. Mueller matrix images of Breast Tissue

After acquiring 16 images, the images are processed in MATLAB platform to acquire intensity component of each and every pixel of image [13]. This Mueller Matrix obtained is normalized to the first element of the matrix to isolate the intensity dependent effects in the image and also simplifies the analysis the Mueller matrix elements are indicated in Table1.

Table1. The	e 16 elemen	ıt Mueller	matrix of	Breast '	Tissue

(M <sub>11</sub> )	(M <sub>12</sub> )	(M <sub>13</sub> )	(M <sub>14</sub> )
1	-0.2791	-0.1860	-0.9535
(M <sub>21</sub> )	(M <sub>22</sub> )	(M <sub>23</sub> )	(M <sub>24</sub> )
0.6977	0.7907	0.5814	1.9767
(M <sub>31</sub> )	(M <sub>32</sub> )	(M <sub>33</sub> )	(M <sub>34</sub> )
0.1628	-0.3488	0.9535	0.0001
(M <sub>41</sub> )	(M <sub>42</sub> )	(M <sub>43</sub> )	(M <sub>44</sub> )
-0.1628	0.7209	0.7442	-0.0465

From the measured Mueller matrix the Diattenuation, Retardance and Depolarization images are acquired and the images are shown in figure 4-6. The mean values of Diattenuation and Depolarization for the sample are shown in Table2.



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Table	2.	Mean	values	OI	Diat	tenua	tion,	Depolar	ization	and

Retardance.					
Property	Value				
Diattenuation	0.8609				
Depolarization	-1.8493				
Retardance	2.2168				



Fig.4. Diattenuation image of Breast Tissue



Fig.5. Depolarization image of Breast Tissue



#### 5. DISCUSSION

The polarization response of Breast Tissue was observed with respect to the extent of polarization variation exhibited by the samples. The diattenuation, depolarization, Retardance values are observed. The depolarization value is due to different scatterings of incident light by sample [10], if the sample surface is lambertian as it exhibits high depolarization of light and thus shows sharp back scattered intensity distribution with high polarization of light. A high specular reflectance nature was observed for the different diattenuations the reason being different surface texture and grain content on the surface though same part of the sample that is used for investigation. Retardance is due to discontinuity present on the sample surface this value is due to compositional and moisture or water content present in the sample as is understood from the Breast tissue composition. Further work is in progress to correlate anisotropic variation in different tissues using the same method.

#### 6. CONCLUSIONS

Optical signature of the Breast tissue sample in the form of a Mueller matrix was obtained and as expected tissue sample exhibited polarization anisotropic character which is evident from the results tabulated. The necessary and sufficient condition for obtaining Complete/Mueller Polarimeter polarization is satisfied which is evident from the images obtained and from the values in the table. The experiment results of this study indicated that pixel by pixel processing of the images obtained effectively identified various optical polarization changes and scattered intensity distributions present in the sample in terms of Diattenuation, Depolarization and Retardance images and values obtained.

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