

Sound as a Qualitative Biospeckle Index Interpretation in the Biospeckle Laser Analysis

Abstract

In this article will be studied two biospeckle analysis methods, in both cases a sound representation of biospeckle analysis will be presented. The first case uses single frequency band model, so that all the spectral information is interpreted by an unique biospeckle index. In the second case a multi frequency band interpretation is used and the signal is separated in N frequency bands, and in these, biospeckle indexes are calculated; finally it is made a sound interpretation of these results. Results of analysis show that these methods are a good [qualitative and relative](#) interpretation of the biospeckle analysis, providing a fast understanding of the state the biospeckle activity in the points analyzed.

Keywords: Biospeckle laser, Biospeckle index, Biospeckle signal, Biological activity, Dynamic speckle, Backscattering.

1. Introduction

The biospeckle laser analysis has presented as a versatile tool in the analysis of biological activity in many types of biological materials; with the grow of the demand in the use of this technique, also are required new methods of present the result. Thus, they are well known in the literature numerical and graphic methods [1, 2], in the case of numerical methods these are a good quantitative form of differentiate between two distinct levels of biospeckle activity, by other side in the case of graphic methods these have showed efficient to show qualitatively the level biospeckle activity in each point of an analysis region to the case of a unique analyzed frequency band, already in the multi frequency band case

the representation of data tends to be more complex to interpret. Thus, born the necessity of design a new form of to show qualitatively the level biospeckle activity, that will be easy to interpret, both in the single frequency band case and in the multi frequency band case. Thereby, in this work is proposed a qualitative biospeckle index interpretation using a sound.

2. System description

The materials used in the tests presented in this work, consist of a laser with a wavelength of 632 nm (red color), pointing to a coffee seed in germinating process, the speckle patten produced is acquired by a digital camera connected to a personal computer with the Octave software [3] installed; additionally was installed over Octave, the biospeckle laser tool library [4, 5]. The acquired images were sampled with a rate of 0.08s, equivalent to a sampling frequency of $F_s = 12.5hz$ into 8 bits format; where, the images have a size of 448×448 pixels (lines and columns respectively), being a total of $M = 128$ images, later the M

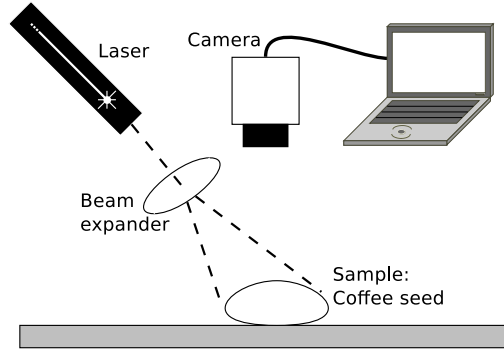


Figure 1: Data acquisition system setup of the coffee seed.

images are grouped in a data package (datapack). Finally, it is in this point that will be apply the tests aforementioned, and biospeckle indexes (BSI) are calculated in accordance with what is explained in the Secs. 3 and 4.

3. Sound interpretation of a biospeckle index

The Fig. 2 represents the sound interpretation system diagram of a biospeckle index; in it, we can see as is selected the i -th point set S_i , from a datapack, and are processed to get any biospeckle index known in the literature [1] [INDEX2] [INDEX3]. Thus, a biospeckle index value BSI_i is obtained from S_i , and this

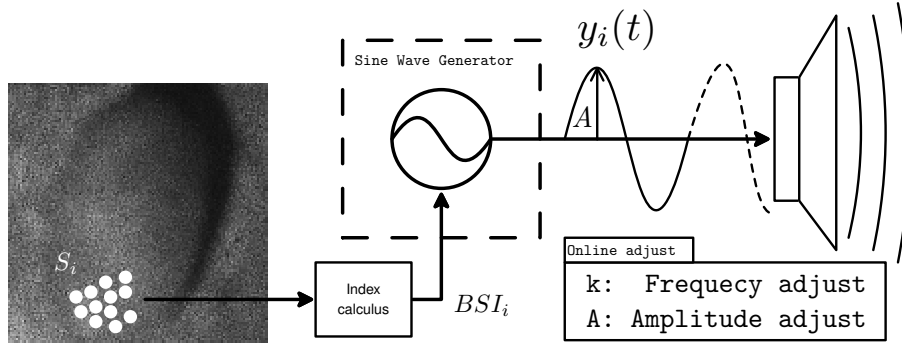


Figure 2: Sound interpretation of a biospeckle index.

information is sent to a sine wave generator that fulfill the Eq. (1),

$$y_i = A \sin(2\pi f_i t), \quad (1)$$

where, A is the amplitude of sound that will be adjusted to suit the user, depending on your hearing level, t is the elapsed time and f_i is the frequency of the wave, that is in function of BSI_i , see Eq. 2.

$$f_i = k f_{ref} \left(\frac{BSI_i}{BSI_{max}} \right) \quad (2)$$

where, BSI_{max} is the maximum expected value of the used index, so that ever $\left(\frac{BSI_i}{BSI_{max}} \right) \leq 1$, f_{ref} is the reference frequency previously predefined by the user thereby f_i ever be a portion of this value, and k is a fine tuning value to f_{ref} which will be adjusted by the user. The last step of the diagram of Fig. 2 shows as the signal y_i is played. It is important to note, that to make the sound signal y_i detected by the human ear, it should be in the audible spectrum [6], this is $20 \leq f_i \leq 20kHz$. But, some people have a reduced audible range, and it is at this point that takes on importance the adjustment by k .

Finally, according the exposed, the Alg. 1 describe the procedure to make a sound interpretation of a *BSI* in a single band system.

Algorithm 1: Sound interpretation of a *BSI* in a single band system.

Data: A datapack *DATA* with M images.

Result: The representation of a biospeckle index through a periodic sound.

- Select the type of biospeckle index to be used;
- Choose the value of f_{ref} ;
- $i \leftarrow 0$;

while *the user need*, **do**

- Select a point set S_i in the datapack *DATA*;
- Calculate the biospeckle index BSI_i over S_i ;
- Evaluate y_i according the Eqs. (1) and (2).
- Play the signal y_i and adjust k and A ;
- $i \leftarrow i + 1$;

if *Do you like exit* **then**

 | break;

end

end

4. Sound interpretation of a biospeckle index in a multi spectral analysis

To present an interpretation of a biospeckle index in a multi spectral analysis, we need first to define the number and positions of the used frequency bands. Thus, the Fig. 3 represents an example case, where 3 frequency bands are used. Given that, the images in datapack were sampled with a frequency F_s ; then, according the Nyquist theorem [7], the maximum signal frequency in the datapack is $F_s/2$. Additionally, we see the values F_j , $\forall j \in \{1, 2, \dots, N\}$, that

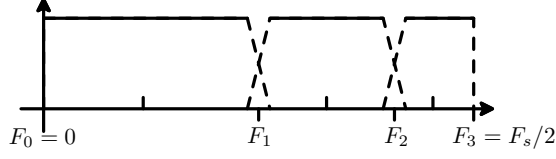


Figure 3: Separation of $N = 3$ frequency bands in a multi spectral analysis.

represent the cut-off frequency bands, being N the number of bands, with the special cases, $F_0 = 0$ and $F_N = F_s/2$.

The Fig. 4 represents the sound interpretation system diagram of a biospeckle index multi spectral analysis, using $N = 3$ frequency bands. Similarly to the

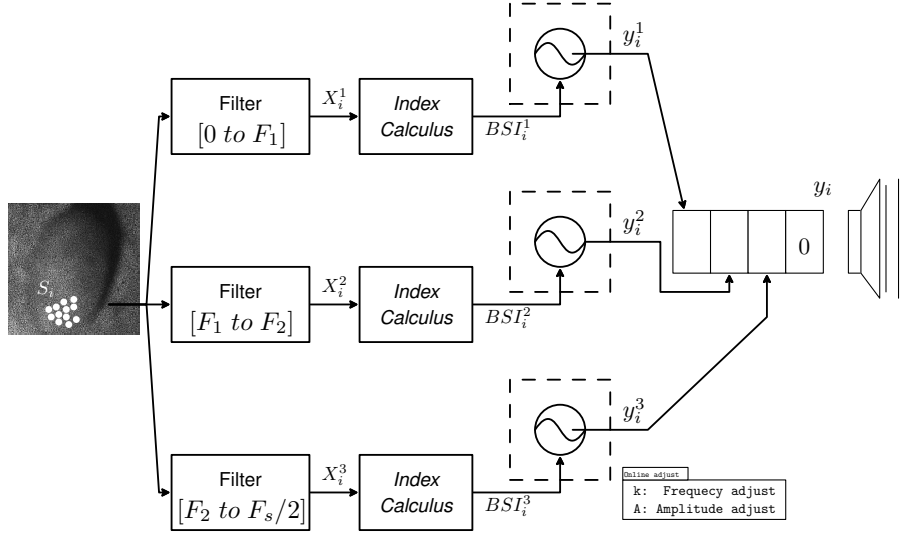


Figure 4: Sound interpretation of a biospeckle index in a multi spectral analysis.

single band case, we can see as is selected the i -th point set S_i , from a datapack; but now, the obtained signal is divided into bands. By example, in the Fig. 4 we have: A low frequency band from 0hz to F_1 , a middle frequency band from F_1 to F_2 , and a high frequency band from F_2 to $F_s/2$; later, over the signal X_i^j , in each band, they are calculated the biospeckle indexes BSI_i^1 , BSI_i^2 and BSI_i^3 , respectively. The indexes are sent to sine wave generators with output

y_i^j as in the Eq. (3),

$$y_i^j(t) = A \sin\left(2\pi f_i^j t\right), \quad (3)$$

and later these signals are grouped in a new signal y_i^j like seen in the Fig. 5; where, i represents the used point set, j represents the used frequency band, A

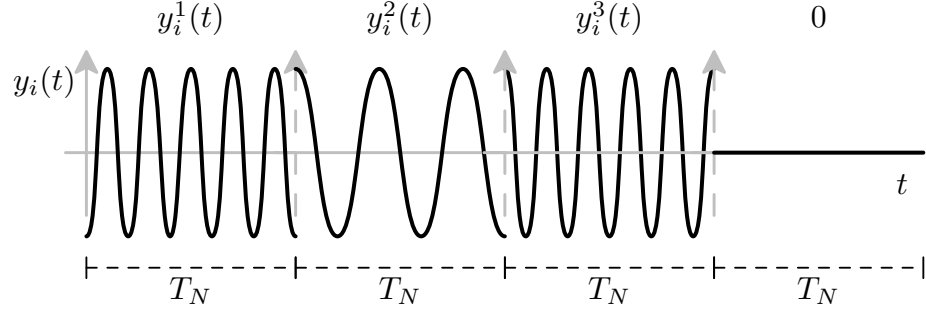


Figure 5: Signal $y_i(t)$ formed by grouping T_N seconds of signals $y_i^1(t)$, $y_i^2(t)$ and $y_i^3(t)$ with T_N seconds of zero samples at end.

is the amplitude of sound that will be adjust to suit the user, t is the elapsed time and f_i^j is in function of BSI_i^j , as can be see in the Eq. (4),

$$f_i^j = k f_{ref} \left(\frac{BSI_i^j}{BSI_{max}} \right), \quad (4)$$

where, BSI_{max} is the maximum expected value of the used biospeckle index in all frequency bands, so that ever $\left(\frac{BSI_i^j}{BSI_{max}}\right) \leq 1$ and f_{ref} is the reference frequency, it should be selected by the user and should be less than $f_s/2$, being f_s the sampling frequency of digital sound synthesizer. Finally the sound signals $y_i^j(t)$ are grouped and the resulted signal $y_i(t)$ is played, see Eq. (5),

$$y_i(t) = \sum_j \Pi(t - (j-1) T_N) y_i^j(t); \quad (5)$$

where, $\Pi(t)$ is a rectangular function of size T_N . The Alg. 2 shows the procedure explained above.

Algorithm 2: Sound interpretation of a BSI in a multi band system.

Data: A datapack $DATA$ with M images.

Result: The representation of biospeckle indexes in a multi band system, through a periodic sound.

- Select the type of biospeckle index to be used;
- Choose the sampling frequency f_s of digital sound synthesizer;
- Choose the cut-off values F_j of the frequency bands;
- $i \leftarrow 0$;

while *the user need*, **do**

- Select a point set S_i in the datapack $DATA$;
- Filter $\forall j$, the information in S_i and return the signals X_i^j ;
- Calculate $\forall j$, the biospeckle index BSI_i^j from X_i^j ;
- Evaluate $\forall j$, y_i^j according the Eqs. (3) and (4).
- Play the signal y_i according the Eq. (5) and adjust k and A ;
- $i \leftarrow i + 1$;

if *Do you like exit* **then**

 | break;

end

end

5. Numerical results

5.1. Numerical results of single band test

To made a single band test will be used the (normalized) *AVD* biospeckle index [1], according the system diagram showed in the Fig. 2. Thus, over the datapack are selected 3 point set (red points, blue points and green points) with 200 points (pixels) each one, being these randomly chosen in the space using a Gaussian probability distribution with radius of 15 pixels, as can be seen in the Fig. 6. To facilitate the selection of points and the interpretation of the

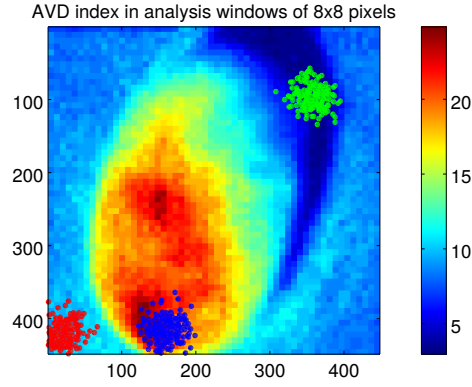


Figure 6: Selecting the points to be analyzed.

results, the point sets were chosen over an image that represent the *AVD* values in graphic mode [4], calculated over all the points (pixels) contained in analysis windows of 8×8 pixels, the relation between the colors and the *AVD* values is expressed by a color bar. Thus, the *AVD* values of the set points are according the Tab. 1, it is easy to see that it match with the data seen in the Fig. 6. Now, according the seen in the Sec. 3 in the Eqs. (1) and (2), the *AVD* biospeckle indexes are interpreted, using the values $A = 1.0$, $k = 4.0$ and $f_{ref} = f_s/2$, being that f_s is the sampling frequency of synthesized sound y_i . Thus, the final sound has a signal with a form as in the Eqs. (6) and (7),

$$y_i = \sin(2\pi f_i t), \quad (6)$$

Table 1: AVD values in the point sets.

	Red points ($i = 1$)	Blue points ($i = 2$)	Green points ($i = 3$)
AVD_i	10.06	21.41	3.70
$AVD_i \frac{2}{255}$	0.079	0.168	0.029

$$\frac{f_i}{f_s} = AVD_i \frac{2}{255}. \quad (7)$$

The Fig. 7 shows, the frequency space behavior of the signals y_i , being the red, blue and green point represented by y_1 , y_2 and y_3 respectively; the graphic was created using the Fourier transform and exemplify as the fundamental frequency of signal y_i change of value in accordance to AVD_i and following the Tab. 1.

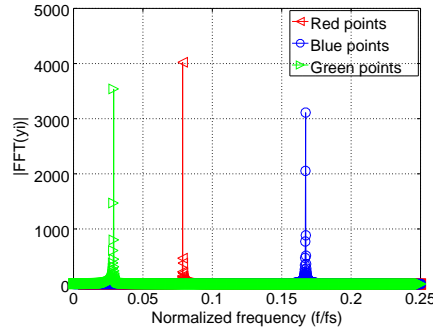


Figure 7: Frequencies of the signals y_i obtained.

The audio files: “*audio-single1.wav*”, “*audio-single2.wav*” and “*audio-single3.wav*”; with the sound interpretations calculated using the Eqs. (6) and (7); from the point sets red, blue and green respectively; they can be downloaded from an on-line repository [FALTA SITE >>>>](#) [8].

5.2. Numerical results of multi band test

To made a biospeckle multi-band test will be used the temporal standard deviation biospeckle index [2], apply to all point set S_i , selected according the

system diagram showed in the Fig. 4, where i is the indexer of selected point set. Thus, over the datapack are selected 3 point sets (S_1 red points, S_2 blue points and S_3 green points) with 200 points (pixels) each one. The same way, that in the single band test, in the multi-band test the points were randomly chosen in the space, using a Gaussian probability distribution with a radius of 15 pixels, see Fig. 8. In this case the selection of points were made over an image that

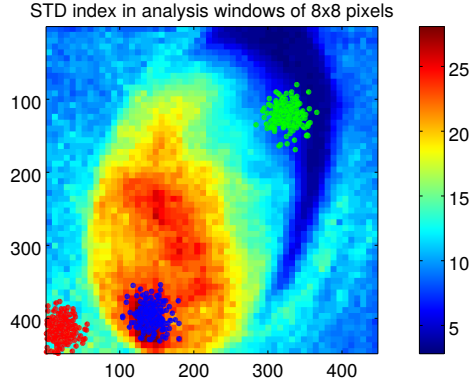


Figure 8: Selecting the points to be analyzed in the biospeckle multi-band test.

represents the temporal standard deviation in graphic mode [4, 2], where it was made with analysis windows of 8×8 pixels; the relation between the colors and the temporal standard deviation biospeckle index values is expressed by a color bar.

As explicated in the Figs. 3 and 4, each point set S_i was separated in 3 frequency bands; thus, in this test, the values of cut-off frequencies will be $F_0 = 0$, $F_1 = \frac{3}{9} \frac{F_s}{2}$, $F_2 = \frac{6}{9} \frac{F_s}{2}$ and $F_3 = \frac{F_s}{2}$, being that the sampling frequency of images in the datapack was $F_s = 12.5 \text{ hz}$. Thereby, the signals X_i^1 , X_i^2 and X_i^3 were obtained from the bands $\{F_0 \rightarrow F_1\}$, $\{F_1 \rightarrow F_2\}$ and $\{F_2 \rightarrow F_3\}$ respectively; over these signals are calculated by each one a temporal standard deviation biospeckle index, being these BSI_i^1 , BSI_i^2 and BSI_i^3 , the values obtained can be seen in the Tab 2.

Finally, using the values $A = 1.0$ and $k = 1.0$ were obtained all the signals y_i^j and these were processed according the Eq. (5) to get the signals y_1 , y_2 and y_3 ,

Table 2: *STD* values in the frequency bands by point sets.

	BSI_i^1	BSI_i^2	BSI_i^3
Red points ($i = 1$)	13.4739	7.3840	6.6311
Blue points ($i = 2$)	20.4776	12.6959	11.8572
Green points ($i = 3$)	5.2657	3.6161	3.5850

representing these to the point sets S_1 , S_2 and S_3 , respectively; as can be seen in the Figure 9. Thus, listening the sound interpretation of the signals y_1 , y_2 and

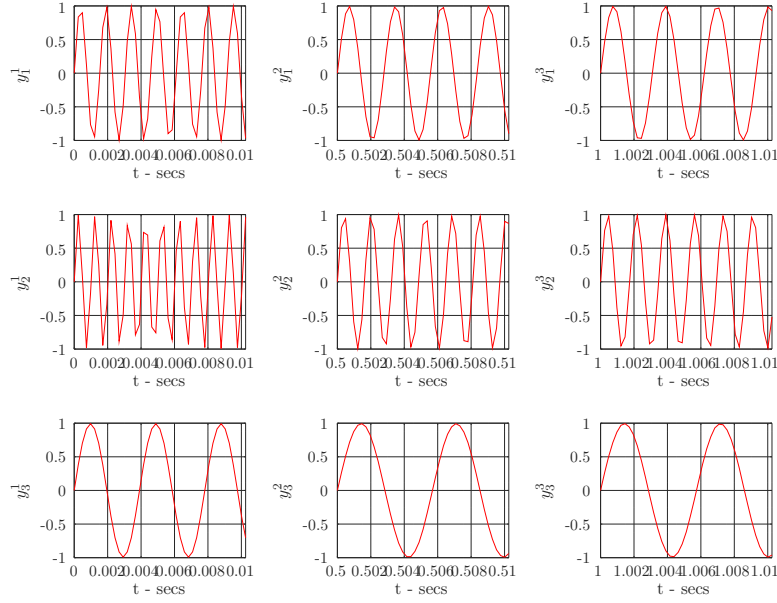


Figure 9: Signals $y_i(t)$ to the 3 point sets.

y_3 , it is easy identify subjectively the level of biospeckle index. The sound files contain these data are the next: “*audio – multi1.wav*”, “*audio – multi2.wav*” and “*audio – multi3.wav*”; they represents the point sets red, blue and green respectively; and they can be downloaded from an on-line repository [FALTA](#)

[SITE](#) > [9]. Additionally, in the same audio files, the 3 frequency bands can be analyzed subjectively the level of biospeckle activity, given the human brain is specialized in differentiating sounds of different frequencies.

6. Distinction of activity level using the biospeckle index

Given that the perception of biospeckle activity using a sound, it is linked to the possibility of distinct between two phenomenon through the comparison between two different levels of the biospeckle index. Then, the next test shows this behavior comparing the *std* biospeckle index of 8 dead seeds with 8 alive seeds, the samples were collected around the radicle of seeds, using 200 points randomly selected using a Gaussian distribution with 15 pixel of radius. In all case the signals were processed, so that each one was subdivide in three frequency bands, like in the Sec. 5.2. Thereby, we obtain the results shown in the Figs. 10, 11 and 12, that represent the *std* biospeckle index of signals in the bands 0 to F_1 , F_1 to F_2 and F_2 to $F_s/2$, respectively.

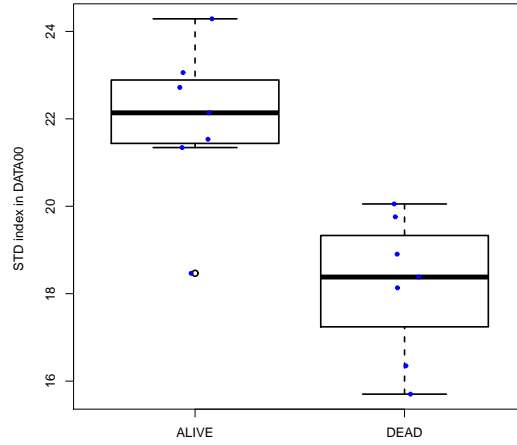


Figure 10: Std index values, of signals between 0 and F_1 hz, in the radicle of 16 dead and alive seeds.

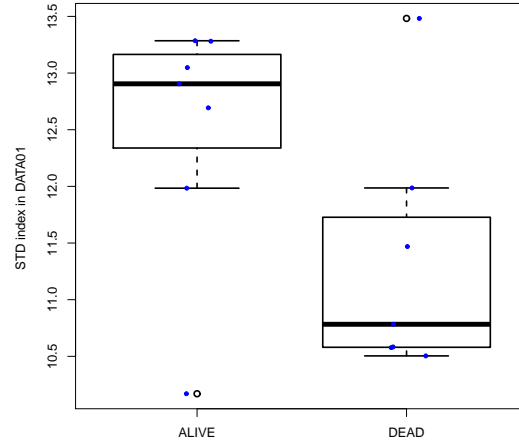


Figure 11: Std index values, of signals between F_1 and F_2 hz, in the radicle of 16 dead and alive seeds.

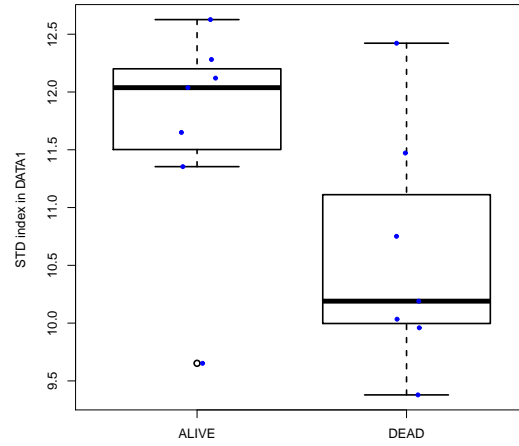


Figure 12: Std index values, of signals between F_2 and $F_s/2$ hz, in the radicle of 16 dead and alive seeds.

It is easy to see that exist different separation levels of the value of biospeckle indexes, depending of the analyzed frequency band; in the test, the band where exist a greater degree of separation it is between 0 and F_1 hz. Given that, an alive seed have a biospeckle index value around 22 and a dead seed a value around 18, it is important to note that it is a statistic analysis and consequently exist samples that not fulfill a pattern, like the sample of alive seed with a lower std value of 18.5, see Fig. 10.

For the above, it is important to note that the technique of to use a sound as a qualitative index, should consider the sensibility level of human ear to distinguish between two frequencies, considering that this configuration also should be influenced by the analyzed frequency band.

7. Conclusion

In this work were presented two methods to the sound interpretation of biospeckle analysis.

The first method analyze the biospeckle signal using a single frequency band model, in this case was showed through of a numerical analysis that the representation using an unique wave form, in this case a sound sine wave, it is efficient; and that the variation of the biospeckle index used can be easily represented and perceived by the frequency variation of sine wave, being it a good tool to a fast biospeckle subjective analysis.

By other side, in the second method, with a multi frequency band approach, it was necessary to assign to each frequency band a different wave form pattern $g^i(f)$, so that the variations of biospeckle index in each band will be linked to repetition frequency variation of the wave form pattern. This method also shown be efficient to identify subjectively the variation of a biospeckle index in a determined frequency band.

8. Acknowledgment

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9. Bibliography

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