

# LASCA-Based Monitoring of Drug Impact and Classification using Machine Learning for Biospeckle Images of Melanoma Cells

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Submitted: 02/05/2024 Revised: 15/06/2024 Accepted: 22/06/2024

**Abstract:** Monitoring a drug's impact on any biological cell is essential for bioengineering and the pharmaceutical field. Preclinical models are available for drug monitoring and testing but are manual and time-consuming. The biospeckle laser technique (BSL) is one optical and nondestructive method that may be useful for measuring cell dynamics. With a deeper comprehension of drug actions and their influence on cellular processes, the BSL approach offers insights into the activity of cell-drug interaction. The technique provides the pattern that the laser beams produce on the screen to look into the activity therein. To Interpret the information using these biospeckle patterns, time and frequency domain algorithms were used in the past. We proposed machine learning and laser speckle contrast analysis LASCA-based analysis of melanoma cells using biospeckle signal/image. Using a machine learning model for computation adds useful inputs to automate the process. In this work, LSACA is used to track the effects of drugs on melanoma cell images. Additional machine learning algorithms are employed to categorize cells in two categories, with and without drug-induced melanoma cells. Various machine learning models are implemented and evaluated using Python language.

**Keywords:** Biospeckle, LASCA, Melanoma cells, machine learning, python

## 1. INTRODUCTION

Preclinical models are used to observe the impact of drugs especially the interaction between cells or tissues, when injected in bio samples. These models provide an understanding of cellular responses and enable the fine-tuning of drug concentrations. To study drug-cell interactions, chemotaxis, investigation of inactive enzymes within living cells, DNA synthesis rate analysis, activation of apoptotic caspases during cell death, and DNA fragmentation techniques have been reported in the literature.

Dynamic laser speckle is an Optical techniques that provide insights into cell-drug interactions[1]. González et al analyzed data numerically and monitored the action of the drug, particularly Colcemid. These methods give a deeper understanding of drug actions and their effects on cellular processes, aiding in the refinement of drug concentrations and treatment strategies before advancing to clinical trials.

Melanoma cells are malignant cells that originate from melanocytes. Melanocytes are the pigment-producing cells in the skin. These cells cause the development of melanoma, a type of skin cancer. Early detection of melanoma is critical for improving treatment and survival rates. Researchers are actively investigating the

biological processes involved in the cause, growth, and spread of skin cancer in the body. Understanding the mechanism of drug interaction with cancerous cells is crucial for developing effective treatments. One promising method for monitoring these interactions is dynamic laser speckle.

Tchivaleva, L et al demonstrated polarization speckle patterns as an important tool for skin cancer detection by conducting a large in vivo clinical study of 214 skin lesions [2].

He demonstrated that statistical moments of speckle patterns are capable of differentiating skin lesions. Three main types of skin cancers are malignant melanoma, squamous, and basal cell carcinoma.

When a surface is irradiated by a laser, the backscattered rays produce a granular structure having temporal fluctuations and phase shifts. This pattern is known as dynamic speckle and if the object is of biological origin this is referred to as biospeckle [3]. This technique allows researchers to monitor, and study various biological processes in real time. By analyzing the dynamic patterns of speckles produced by the interaction of laser light with the sample, it is possible to gain insights into cellular activities and the effects of drug treatment. Biospeckle patterns are obtained due to microscopic movements and refractive index variation in biological sample. This is analyzed using statistical analysis, processing in the frequency domain, and processing in the time domain [4]. Different types of signal descriptors and algorithms were used by researchers for analyzing activity such as Spatial contrast, speckle time evolution, time history of the

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speckle pattern (THSP)[5], occurrence matrix (COM), Fujii’s method, structural function, motion history image and many more.[6].

The inclusion of emerging technologies artificial intelligence for data /signal analysis, automates the process of analysis and reduces the computational time. The use of automated image analysis techniques for optical metrology has seen significant growth in the last few years [7], [8]. Researchers have analyzed & classified the speckles by computational methods for seed classification, and medical and biometric-based applications.

Melanoma is a skin cancer that occurs in melanocytes. This produces melanin that gives skin color. This type of cancer most commonly occurs in the skin but it can also develop in other body parts containing melanocytes. The paper includes the following sections: theory, proposed method, results, and conclusion. Proposed method and results, Illustrate in-depth implementation details of the model and the analysis of results. In the last section concluding remarks are reported.

## 2. THEORY

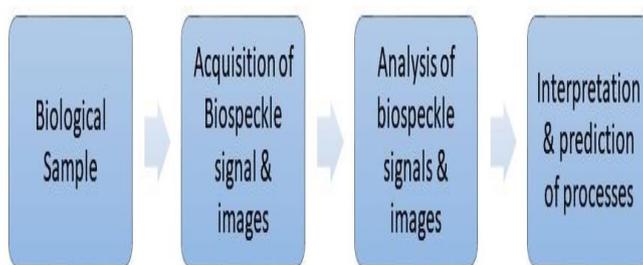
The proposed work is based on the processing of signal/ images acquired by Laser biospeckle technique and use of computational model for analysis purpose.

Biospeckle signal/images are dynamic speckle patterns generated by biological samples when illuminated with coherent laser light. This technique, also known as dynamic speckle, has utility in various domains such as

Exposure to ultraviolet (UV) radiation from sunlight or tanning beds causes DNA damage to the skin and results in Melanoma disease. González et al proposed a biospeckle technique for the monitoring of drug impact on melanoma cells [1].

In this paper, melanoma cell activity is monitored using biospeckle data available on the cloud. We presented the classification technique to classify drug-induced melanoma cells and diseased melanoma cells using biospeckle images. For monitoring and classification, machine learning (ML) models are deployed on image data. A detailed review of bast analysis using ML was presented by Ghaderzadeh et al for the detection and classification of blood smears[9]. The Author reported the common features that can be extracted and used for image analysis.

agriculture, biology, medicine, and food science. Biospeckle signals and image acquisition are the first steps for the analysis of activity via the biospeckle technique. It is used to monitor activity in the sample of biological origin The biospeckle signals and images are acquired via experimental setup, synthetic simulation techniques, and open-source resources. The biospeckle laser (BSL) experimental setup consists of the laser source, CCD camera, beam expander, and computing device/algorithms [10]. When an object is illuminated with a laser and the surface is rough concerning the radiation wavelength, interference between many random paths of various lengths produces speckles. The steps involved in the technique can be summarized in Figure 1:



**Fig 1** Steps involved in biospeckle technique-based application

Laser speckle contrast analysis (LSACA) is used for monitoring drug impact on biospeckle images of melanoma cells. LASCA is based on spatial or temporal contrast. The speckle contrast is a measure of the variation in intensity of the speckle pattern over time. The speckle contrast  $C$  is calculated for each pixel or region of the captured images. Speckle contrast is the ratio of the standard deviation to the mean intensity of the speckle pattern over a small temporal window and is given by equation 1.

$$\text{Speckle contrast } C = \sigma / I \quad (1)$$

$\sigma$  and  $I$  is the standard deviation of the spatial intensity variations and the ensemble average of the intensity. It provides an activity map of the area of interest in real time without scanning[11]. This method is used to monitor the impact of drugs on cancerous cells.

**2.2 Computational model:** Machine Learning. (ML) is a branch of artificial intelligence, consisting of algorithms and mathematical relations. These algorithms and mathematical relations used in clinical research and other applications for monitoring and classification. ML is a

powerful approach for analyzing medical images and aiding in diagnoses. The relevant feature selection is an important aspect of the deployment of any ML model. This process involves identifying aspects of the images to distinguish between classes, for example, healthy and diseased tissue. The machine learning algorithm then learns patterns from these features to make classifications. Machine learning algorithms are classified into supervised, unsupervised, Semi-supervised reinforcement, and ensemble learning algorithms. Decision Trees, Random Forest, k-nearest neighbor's and support vector machine (SVM) algorithms are used in supervised learning whereas Principal Component

Analysis (PCA), K-Means Clustering, Hierarchical Clustering and autoencoders used in unsupervised learning. The algorithm performance is evaluated by determining accuracy, sensitivity, specificity, recall, and precision. These metrics provide insights about the effectiveness of algorithm in a clinical application and environment. The selection of the algorithm depends on the nature of the data, the application, computational resources, and the desired level of performance.[12].

### 3. PROPOSED METHOD

This section describes the methodology (Figure 2) of work presented here.

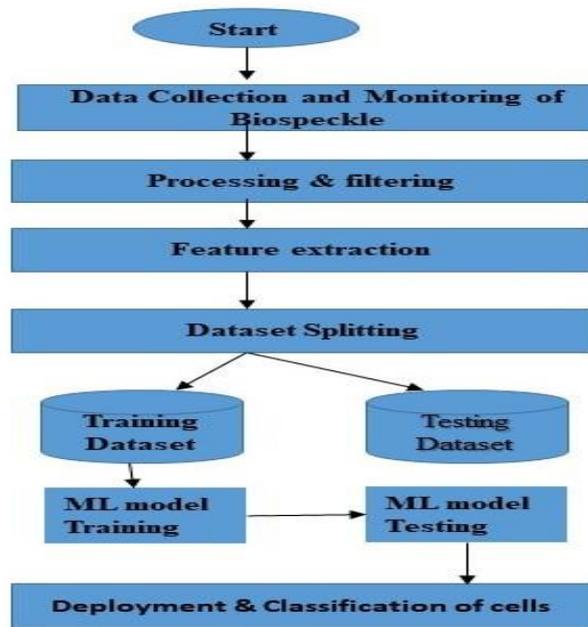


Fig 2 Methodology

#### 3.1 Data Collection and Monitoring of Biospeckle

Biosepeckle images of melanoma cells and drug-induced melanoma cells collected from open source platform biospeckle laser on cloud [13]. The dataset consists of three

types of biospeckle images one group belongs to melanoma cell images. The other group consist of melanoma cell images having Colcemid drug concentrations of 0.4  $\mu\text{g}/\text{mL}$  and 0.2  $\mu\text{g}/\text{mL}$ . Sample images of melanoma cell and drug induced melanoma cell are shown in Figure 3.

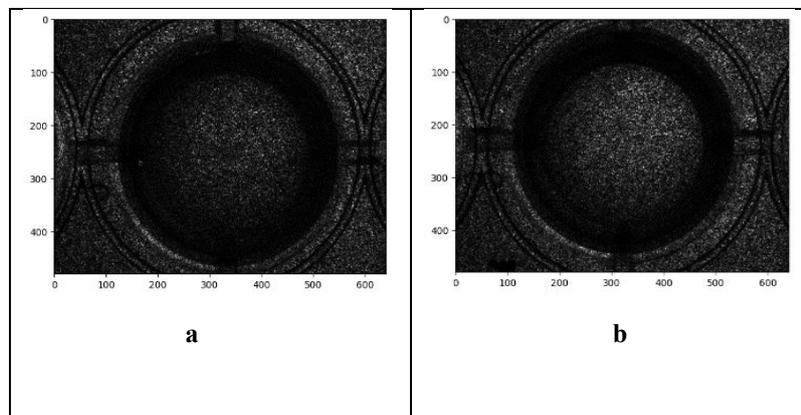


Fig 3 a. Melanoma cell, b. Drug-induced Melanoma cell

To investigate cell activity in the biospeckle images LASCA algorithm is applied to obtain a spatial map of speckle contrast. Laser light scattered from a biological cell produces a speckle pattern. The change in the speckle pattern is related to the velocity of the movement. LASCA generates spatial maps of speckle contrast, which represent the distribution of blood flow or cell motion within the imaged area. Spatial features, such as spatial frequency content, entropy, or spatial autocorrelation, are extracted from these maps to characterize the spatial distribution of blood flow or tissue motion and identify regions of interest. Initial cell motion is measured by melanoma cells to establish a reference. After drug administration cell motion is measured at various intervals to observe changes. Variations in speckle contrast values are analyzed to assess the impact of the drug treatment on cancerous melanoma cells.

**3.2 Data processing & filtering:** The dataset consists of a total of 384 images. The size of the image dataset is increased with data augmentation techniques such as flipping, color adjustments, random cropping, and rotations. The collected images are converted to greyscale and pre-processed by contrast stretching, plotting histogram, Otsu's thresholding, and filtering are applied to images. Filtering is used to remove unwanted information.

**3.3. Feature extraction:** Program code developed in Python language, OpenCV and skimage library used for the feature extraction. Different features such as area, aspect ratio, extent, bounding box, mean and standard deviation, contrast, entropy, local binary features, and image moments, Time history speckle pattern, occurrence matrix are extracted from biospeckle data. All extracted attributes are saved in a CSV file for further analysis.

**3.3.4 Dataset splitting:** The CSV file with the attribute of all dataset images is loaded. For Testing and training purpose, the original dataset is splitted into 1:3 ratio. Randomly 25% of image data is selected as test data and used for model testing. The remaining data is used for model training. Both folders have images of two classes namely melanoma cells & drug-induced melanoma cells.

**3.3.5 Model Training & Testing:** For the model training and testing different models deployed on the extracted .csv extension file. This section describes the models[14,15] used to classify biospeckle images of melanoma cells in two classes, with and without drug. Keras is used in conjunction with the TensorFlow backend to construct the model. The following algorithms were deployed for the model training and testing:

Gaussian naïve bayes (GNB): This model is based on Bayes' theorem to calculate the probability of a certain class based on features. In the training phase, the algorithm estimates the mean and variance of each image for classification.

Support vector machine (SVM): SVM is based on finding the hyperplane that best separates the data points of different classes. SVM uses kernel functions to transform data. The radial Basis Function (RBF) kernel is used for training and fitting. This kernel gives high accuracy and takes less time.

k-nearest neighbor's algorithms: An input feature is classified using the k-nearest neighbors' model by allocating the object to the most comparable class. The unknown object is simply placed in the class of that one nearest neighbor if  $k = 1$ . The Euclidean distance is the measure of an object's proximity to another, as determined by the similarity function.

Decision tree model (DT): DT recursively split the data into subsets based on the features. These features provide the best separation. It is based on criteria like Gini impurity and information gain. This process repeated till the data is perfectly classified or a stopping criterion is met.

Random forest model: Random forest can be categorized as a supervised ML, where the decision-making is performed using the N-Decision tree approach. This can be mathematically expressed as:

$$E \sum_{i=1}^c -p_i \log_2 p_i \quad (2)$$

where, E=entropy

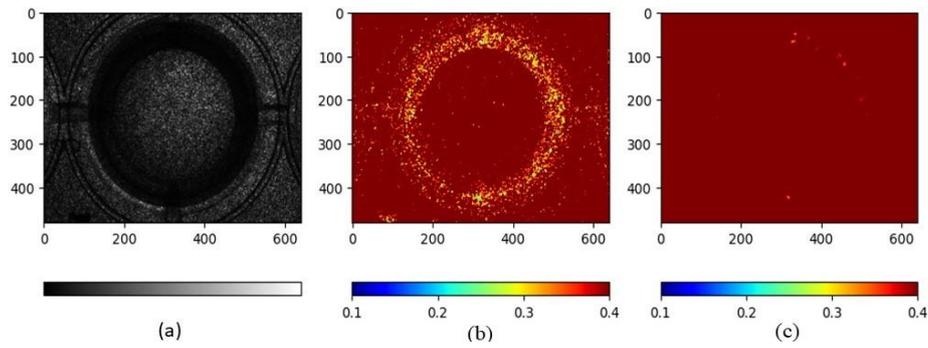
$p_i$ =probability of occurrence of an event i

c=total number of events

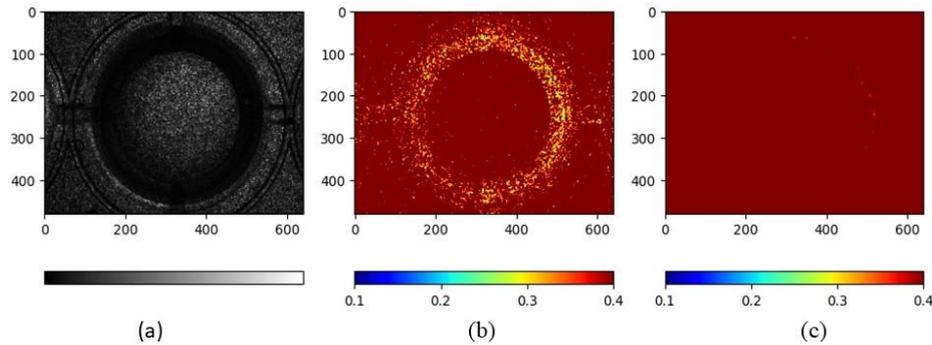
## 4. Results

Laser speckle contrast analysis is a full-field technique for analyzing activity occurring inside the cancer cells. For contrast analysis, a 5\*5 and a 10\*10 window are used over BSL images. Figure 4 presents the original biospeckle (BSL) image of the cell and its contrast map for both window sizes.

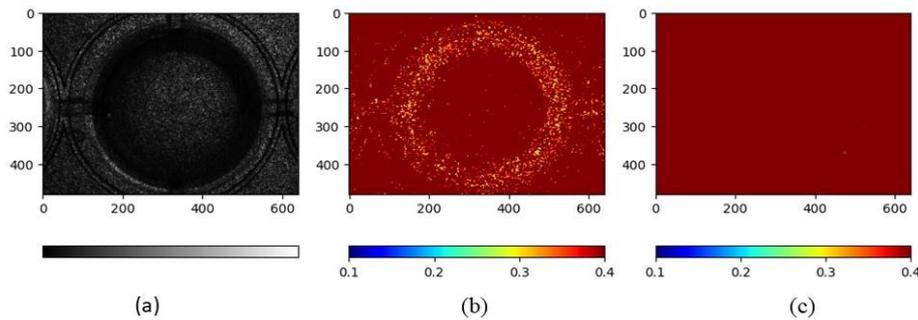
The dataset consists of biospeckles of Colcemid drug-induced melanoma cells in two different concentrations [2]. Figures 5 & 6 visualize the LASCA images of drug-induced cell.



**Fig 4** LASCA Images without drug a) BSL Image b) contrast map using 5\*5 window c) contrast map using 10\*10 window



**Fig 5** LASCA Images with drug concentration 0.2 µg/mL a) original BSL Image b) contrast map using 5\*5 window c) contrast map using 10\*10 window

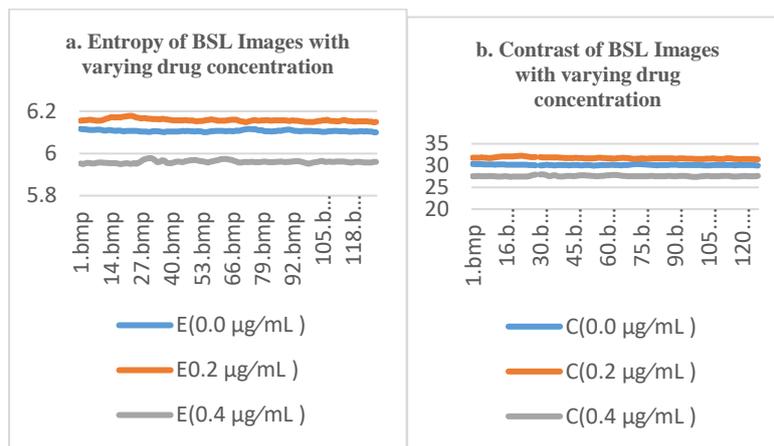


**Fig 6** LASCA Images with induced drug concentration 0.4 µg/mL a) original BSL Image b) contrast map using 5\*5 window c) contrast map using 10\*10 window

LASCA images in figure 5,6 are illustrative of blood flow changes, in the melanoma cell after inducing the drug in varying concentrations. The speckle contrast map is generated using 5\*5 And 10\*10 window size.

Active cell metabolism, rapid growth, or movement of microorganisms cause intensity variations in speckle images. Based on intensities, contrast, and entropy values are computed. High entropy and contrast values in bio-speckle images indicate fast biological activity or

movement within the sample. Figure 7 illustrates the comparative analysis of feature variations for different time series biospeckle images. It is visible that when the drug of low concentration 0.2 µg/mL is induced in the cell higher entropy and contrast values are obtained than the cell without the drug. Further, as the concentration of the drug increased to 0.4 µg/mL entropy and contrast, both values get reduced. This indicates that with an increase in drug concentration, cell activity is reduced significantly.



**Fig 7** Feature variation in time series biospeckle images with varying drug concentration a. Entropy b) contrast

The first step of ML model deployment is data pre-processing and then features are extracted for model training and testing. Spatial and temporal features mean, standard deviation, contrast, entropy, inertia moment (IM), and Local Binary Patterns LBP are extracted from each image. ML algorithms Gaussian Naive Bayes

(GNB), k-nearest neighbor (KNN), support vector machine (SVM), decision tree (DT), and random forest (RF) are used for training.

The performance of each ML model is evaluated and shown in Table 1.

**Table 1:** Performance Parameters of machine learning (ML) model

Model	Accuracy	Precision	Recall
GNB	80.11	79.67	90.25
SVM	90.25	96.82	93.29
KNN	91.86	90	90.82
DT	94.77	93.37	96.77
RF	94.77	98.37	96.77

GNB gives the lowest accuracy and precision relative to other models. Its recall value indicates that GNB is identifying positive instances effectively. SVM performance is well in terms of precision and it minimizes the false positives effectively. DT performance is balanced across all metrics. RF provides balanced performance, with a slightly higher precision than DT. For tasks prioritizing precision, RF and KNN are the best-suited models. For balanced performance, DT and RF-based models are preferred. Further to simulate the behavior of the ML model for noisy data, a synthetic data set was created and tested for each model. Speckle noise is added to the melanoma test data set and its performance is also evaluated. For noisy data sets, it is observed that the performance of the model degrades. The accuracy, precision, and recall are 49.23, 48.36, and 45.26 in the case of noisy data.

## 5 CONCLUSION

The presented work illustrates the use of LASCA and machine learning models for tracking the impact of drug

on melanoma cells. The dataset comprised of biospeckle images of melanoma cells treated with different concentrations of Colcemid drug. Biological activity inside the cells is demonstrated based on various spatial and temporal characteristics. This study analyses biospeckle images of melanoma cells, with and without drugs using the LASCA method. The LASCA contrast images demonstrate that activity inside of melanoma cells decreases with an increase in colcemid drug concentration exceeding threshold value. In addition to activity analysis, machine learning models, SVM, KNN, RF, and GNB deployed for performance evaluation against both the standard test data and a noisy dataset. It is observed that model performance degrades in the presence of noise. Amongst all deployed models, optimum performance is achieved with the model.

In brief, this work highlights the potential of LASCA and machine learning for monitoring and classifying melanoma cells with drug and without drug. It opens up scope for the further research on real-time image data to

automate the classification and monitoring process for medical & clinical research domains. The findings elaborate that the models perform well on clean data. However, the performance degrades with the noisy data as there is reduction in performance parameters for synthetic noise data.

**Competing Interests:** Not Applicable

**Funding Information:** Not Applicable

**Author contribution:** Not Applicable

**Data Availability Statement:** Not Applicable

**Research Involving Human and /or Animals:** Not Applicable

**Informed Consent:** Not Applicable

## ACKNOWLEDGMENT

Authors gratefully acknowledge Gonzalez-Peña and Rolando J. from Univ.de Valencia (Spain), Univ Federal de Lavras (Brazil) for providing biospeckle images on open-source platform “Biospeckle laser on clouds “

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