

# Cardiac Phase Retrieval in Intravascular Ultrasound Sequences

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**Abstract:** - Acquisition of intravascular ultrasound (IVUS) sequences is a major artifact for 3-D exploring of coronary arteries. Most current techniques are based on the electrocardiogram (ECG) signal to obtain a gated pullback without longitudinal motion by using specific hardware or the ECG signal itself. We present an image-based approach for cardiac phase retrieval from coronary IVUS sequences without an ECG signal. A signal reflecting cardiac motion is computed by exploring the image intensity local mean evolution. The signal is filtered by a series of filters namely Gaussian and Optical at the main cardiac frequency. After applying minimum and maximum values of threshold, based on the pixel values at different position, the cardiac phase is retrieved and the report is mailed to the concerned.

**Keywords:** ECG, IVU; Image intensity; Gaussian filter; Optical filter; Cardiac phase.

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## I. INTRODUCTION

The heart is a muscular organ about the size of a fist in both humans and other animals, which pumps blood through the blood vessels of the circulatory system. The diseases which involve heart and blood vessels are known as cardiovascular diseases (CVD). These diseases are caused by high blood pressure, smoking, lack of exercise, obesity, high blood cholesterol and poor diet etc. There is general consensus that cardiovascular diseases (cerebrovascular, peripheral arterial, coronary heart and strokes among others) are a leading cause of death. According to the World Health Organization, they constitute approximately a 30% of all global deaths and they are projected to remain the single leading causes of death. Among cardiovascular diseases, coronary heart pathologies (which can trigger anginas and heart strokes, for instance) constitute the gross of these deaths with a 33%. Artery diseases are mainly caused by the accumulation of plaque (made up of a combination of blood cholesterol, fat and cells) inside arterial walls. Such plaque accumulation narrows the artery's blood flow (stenosis) and makes arteries inflaming and being less flexible (atherosclerosis). Artery blood flow reduction is measured by the percentage of obstruction in vessel sections and is a usual measurement previous to decide which is the best treatment (either surgical or pharmacological) for an atherosclerotic lesion. Depending on the histological composition of the plaque, its (bio-mechanical) physical behavior will be different, making it more or less unstable (vulnerable plaques) and, thus, resulting in a different risk for the patient. Early detection of plaque composition is a main step for planning the most suitable treatment (angioplasty, stent apposition) and might prevent further thrombosis potentially leading to a fatal heart attack. Tissue bio-mechanical properties play an important role in the diagnosis and treatment of cardiovascular diseases. The main mechanical properties currently under study are radial strain, which is related to plaque type and vulnerability, and shear stress, which influences the probability of plaque accumulation. Both measures can be computed by means of

the study of vessel tissue deformation along the cardiac cycle [1]–[4].

Longitudinal motion during in vivo pullbacks acquisition of Intra Vascular Ultra Sound (IVUS) sequences is a major artifact for 3-D exploring of coronary arteries. Most current techniques are based on the electrocardiogram (ECG) signal to obtain a gated pullback without longitudinal motion by using specific hardware or the ECG signal itself. We present an image-based approach for cardiac phase retrieval from coronary IVUS sequences without an ECG signal. A signal reflecting cardiac motion is computed by exploring the image intensity local mean evolution. The signal is filtered by a series of filters namely Gaussian and optical, centered at the main cardiac frequency. Thresholding is applied to get the maximum and minimum values and heart rate is calculated with the help of pixel values at different location through which condition of the heart can be decided.

Existing strategies for image-based gating follow these steps. First, a signal reflecting cardiac motion is extracted from IVUS sequences. Second, the signal is filtered to remove non-cardiac phenomena and artifacts. Finally, a suitable sampling of the filtered signal is used to retrieve cardiac phase. All authors agree on using the extrema of a signal reflecting cardiac motion for sampling it at the same part of the cardiac phase. The main differences among existing algorithms and, thus, the clue for accurate cardiac phase retrieval, are on the computation of the cardiac signal and its further filtering.

Cardiac phase is obtained by exploring temporal changes of either vessel structures (lumen size) [5], [6] or image gray-level [7], [8], [9]. Given that segmentation of IVUS images is not straightforward, exploring vessel structures requires manual intervention. Compute the changes of lumen size by means of the area enclosed by a manual segmentation of vessel lumen. Explore lumen size evolution by computing gray-level statistics inside a manually defined ROI. Image intensity methods do not require user intervention, because they do not need identification of vessel structures. However, the choice of the similarity measure detecting changes between consecutive frames is a delicate procedure.

## II. RELATED WORK

Aura Hernández, David Rotger and Debora Gil (2008) have proposed a longitudinal motion artefacts in Intravascular Ultrasound (IVUS) sequences hinders a properly 3D reconstruction and vessel measurements. Most of current techniques based on the ECG signal to obtain a gated pullback without the longitudinal artifact by using a specific hardware or the ECG signal itself. The potential of IVUS images processing for phase retrieval still remains little explored. In this paper, they have presented a fast forward image-based algorithm to approach ECG sampling. Inspired on the fact that maximum and minimum lumen areas are related to end-systole and end-diastole, their cardiac phase retrieval is based on the analysis of tissue density of mass along the sequence. The comparison between automatic and manual phase retrieval (0:07 + 0:07 mm. of error) encourages a deep validation contrasting with ECG signals.

Hernández-Sabaté<sup>1</sup>, M.M.S. Matsumoto<sup>2</sup>, S.S. Furuie<sup>2</sup>, D. Gil<sup>1</sup>(2010) have proposed a good reliable approach to cardiac triggering which is of most importance in obtaining accurate quantitative results of atherosclerotic plaque burden from the analysis of Intravascular Ultrasound sequences. Although, in the last years, there has been an increase in research of methods for retrospective gating, there is no general consensus in a validation protocol. In this paper, they propose an objective validation protocol based on the variability of the retrieved cardiac phase and explore the capability of several quality measures for quantifying such variability. They noticed that the residual variance of the regression correlation line is robust against fraction and variability's as far as one can establish a pair-wise correspondence between candidate and reference.

Bourantas, Plissiti, Rees and K Michalis (2005) have proposed a evaluation of a new semi-automated intravascular ultrasound (IVUS) border detection method. The method was used to identify the lumen and the external elastic membrane or the borders of stents in 80 IVUS images, randomly selected from 10 consecutive human coronary arteries. These semi automated results were compared with observations of two experts. Several indices in each case were obtained in order fully to evaluate the method. The time required for identification of the borders was also recorded. The inter observer variability of the method ranged from 1.21% to 5.61%, the correlation coefficient from 0.98 to 0.99, the slope was close to unity (0.94–1.03), the y intercept close to zero and the Williams index value was close to unity (range 0.67–0.91). The time required for the method to identify the borders of the different vessel layers for the whole IVUS sequence was 5.2±0.2 min. The results demonstrate that the method is reliable and capable of identifying rapidly and accurately the different vessel layers depicted in IVUS images

Kay, Wardeh, K. Kozuma, G. Sianos, E. Regar, and M. Knook,(2001) have proposed a thesis where they worked on ECG-gated intravascular ultrasound with three-dimensional reconstruction which was performed post stent implantation and at the 6-month follow-up to assess restenosis within the

margins of the stent and at the stent edges in 16 patients. Angiographic restenosis was witnessed in four patients, all in the proximal in-stent position. By intravascular ultrasound in-stent neointimal hyperplasia, with a >50% stented cross-sectional area, was seen in eight patients. This was witnessed proximally (n=2), distally (n=2) and in both segments (n=4). Echolucent tissue, dubbed the 'black hole' was seen as a significant component of neointimal hyperplasia in six out of the eight cases of restenosis.

Gozde Gul Isguder, Gozde Unal, Martin Groher, Nassir Navab, Ali Kemal Kalkan, and Johannes Rieber (2008) have proposed a imaging technology which provides cross-sectional images of internal coronary vessel structures. The IVUS frames are acquired by pulling the catheter back with a motor running at a constant speed. However, during the pullback, some artifacts occur due to the beating heart. These artifacts cause inaccurate measurements for total vessel and lumen volume and limitation for further processing. Elimination of these artifacts are possible with an ECG (electrocardiogram) signal, which determines the time interval corresponding to a particular phase of the cardiac cycle. However, using ECG signal requires a special gating unit, which causes loss of important information about the vessel, and furthermore, ECG gating function may not be available in all clinical systems. To address this problem, they proposed an image-based gating technique based on manifold learning.

Aura Hernández-Sabaté, Debora Gil, Eduard Fernandez-Nofrerias, Petia Radeva, and Enric Martí (2009) have declared that Tissue biomechanical properties (like strain and stress) are playing an increasing role in diagnosis and long-term treatment of intravascular coronary diseases. Their assessment strongly relies on estimation of vessel wall deformation. Since intravascular ultrasound (IVUS) sequences allow visualizing vessel morphology and reflect its dynamics, this technique represents a useful tool for evaluation of tissue mechanical properties. Image misalignment introduced by vessel-catheter motion is a major artifact for a proper tracking of tissue deformation. In this work, they focus on compensating and assessing IVUS rigid in-plane motion due to heart beating. Motion parameters are computed by considering both the vessel geometry and its appearance in the image. Continuum mechanics laws serve to introduce a novel score measuring motion reduction in vivo sequences. Synthetic experiments validate the proposed score as measure of motion parameters accuracy; whereas results in vivo pullbacks show the reliability of the presented methodologies in clinical cases.

Hui Zhu, Kevin D. Oakeson, and Morton H. Friedman (2003) have proposed most of the quantitative measures from Intravascular Ultrasound (IVUS) images which vary with the cardiac cycle. Although ECG-gated acquisition can prevent the pulsations from influencing the measurements, it may extend the acquisition time, and furthermore, very few IVUS systems currently in clinical use incorporate ECG-gated function. In this paper, they have presented a practical method to retrieve cardiac phase information directly from in vivo clinical IVUS image sequences. In an IVUS image

that contains a cross-section of coronary artery, there are three regions annularly distributed from the center of the image – catheter, lumen, and part of the vessel wall. The catheter region exhibits virtually no change from frame to frame during the catheter pullback. While the lumen is a dark region, the vessel wall region appears bright. The change in lumen size and position that accompanies the pulse causes the image intensity of the IVUS images to exhibit a periodic variation along the pullback path. By extracting this signal attributed to the cardiac cycle, a subsequence of frames during pullback at the same phase of the cardiac cycle can be selected. The method was tested by the IVUS images of both a coronary phantom and a patient.

Monica M. S. Matsumoto<sup>a, b</sup>, Pedro Alves Lemos<sup>b</sup>, Takashi Yoneyama<sup>a</sup>, Sergio Shiguemi Furuieb (2008) have presented a framework to prove Image gating is related to image modalities that involve quasi-periodic moving organs. Therefore, during intravascular ultrasound (IVUS) examination, there is cardiac movement interference. In this paper, they aimed to obtain IVUS gated images based on the images themselves. This would allow the reconstruction of 3D coronaries with temporal accuracy for any cardiac phase, which is an advantage over the ECG-gated acquisition that shows a single one. It is also important for retrospective studies, as in existing IVUS databases there are no additional reference signals (ECG). From the images, we calculated signals based on average intensity (AI), and, from consecutive frames, average intensity difference (AID), cross-correlation coefficient (CC) and mutual information (MI). The process includes a wavelet-based filter step and ascendant zero-cross detection in order to obtain the phase information. Firstly, we tested 90 simulated sequences with 1025 frames each. Their method was able to achieve more than 95.0% of true positives and less than 2.3% of false positives ratio, for all signals. Afterwards, they tested in a real examination, with 897 frames and ECG as gold-standard. They achieved 97.4% of true positives (CC and MI), and 2.5% of false positives. For future works,

methodology should be tested in wider range of IVUS examinations.

Francesco Ciompi, Carlo Gatta, Oriol Pujol, Oriol Rodríguez-Leor, Josepa Mauri Ferré, and Petia Radeva (2006) have proposed this paper where atherosclerotic plaque has been identified as one of the most important causes of sudden cardiac failure in patients with no history of heart disease. Intravascular Ultrasound (IVUS) represents a unique technique to study, determine and quantify plaque composition and thus allows to develop automatic diagnostic and prediction techniques for coronary diagnosis and therapy. However, one of the main problems of image-based studies is its dependence on image brightness and data miss-registration due to the dynamic system composed by the catheter and the vessel. Hence, the high dependence of the automatic analysis on the gain setting of IVUS console and its transmit power as well as vessel motion make impossible direct analysis, comparison and follow up of IVUS studies. To this purpose, a complete framework for data analysis should be considered focusing on: a) modeling the image acquisition and formation process, b) developing techniques for removing data acquisition artifacts due to the nature of ultrasound reflectance and motion of coronary vessels, c) developing sophisticated tools for extracting features from radio-frequency and images, and d) designing robust methods to discover and classify different categories of tissue structures. From the literature survey it is observed that very less work is cited on cardiac phase retrieval based on video sequences. Hence the proposed work is performed.

### III. PROPOSED SYSTEM

The proposed system contains the following modules as shown in “fig 1”

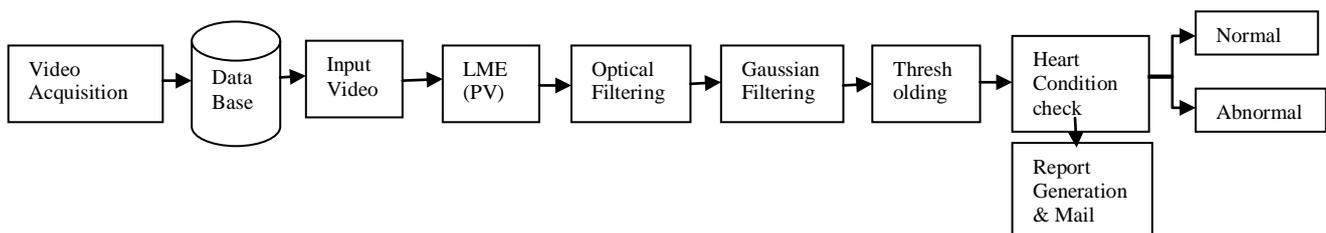


Fig 1: Proposed System Design

#### 3.1 Video Acquisition

Video acquisition in image processing can be broadly defined as the action of retrieving a video from some source, usually hardware based source, so it can be passed through whatever process need to occur afterwards. Performing video acquisition in image processing is always a first step in the workflow sequence because, without an image no processing is possible. One of the ultimate goals of video acquisition in image processing is to have a source of input

that operates within such controlled and measured guidelines. Here the video is obtained by a hardware device called catheter device which is placed in the heart to get the IVUS video sequence which are in turn stored in database.

#### 3.2 Database

Database is a collection of related data, here the data is meant to be IVUS video samples. There are 6 IVUS video samples that have been collected to carry out the proceedings to achieve the expected results.

### 3.3 Input Video

Here in this module one should select the video from the database of IVUS video samples and pass it to the next module for further processing of the video.

### 3.4 Local Mean Evolution (Pixel Variation)

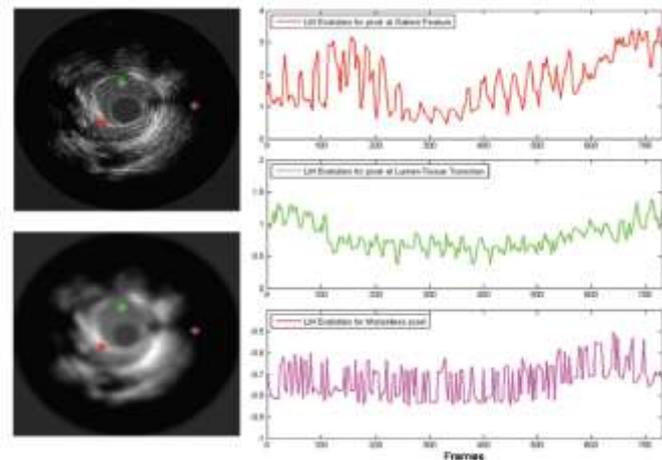


Fig.2 Local mean evolution of different pixels. On the left, the original intravascular ultrasound sequence (top image) and the one corresponding to local mean (bottom image). On the right, the graphics of the evolution of three pixels, a pixel at a salient feature (top), a pixel at lumen-tissue transition (middle), and a motionless pixel (bottom).

Fig. 2 shows LM evolution for three different pixels from left to right, a pixel on a calcified plaque (identified in red), a pixel at luminal cavity close to lumen-vessel interface (identified in green), and a motionless pixel (identified in magenta). Images on the left-hand side show the original IVUS sequence (top image) and the one corresponding to LM (bottom image). The positions of the three pixels are marked with asterisks. Points reflecting cardiac motion correspond to the projection of such points onto the image plane (usually the first sequence frame). The graphs on the right side of Fig. 2 plot LM profiles along the pullback for (from top to bottom) calcified plaque (longitudinal) transition, lumen-vessel interface, and motionless pixels. The red pixel on the calcified plaque reflects longitudinal motion by the 3-D transition between calcified and non-calcified structures along the pullback (the plaque crosses the position of the red pixel because of cardiac motion). This is reflected in the top plot on the right-hand side. The green pixel is close to the lumen wall interface, though located inside the luminal cavity. Therefore, in the original IVUS sequence, the lumen-wall interface does not cross its position at any frame. This is reflected in a periodic pattern (although weaker than the calcium pixel) in the middle LM plot on the right-hand side. Finally, the motionless pixel located in a background shadow does not reflect any persistent pattern in any of the sequences (original or LM).

### 3.5 Optical Filtering

The impact of background noise and textured areas, as well as non-cardiac dynamic phenomena (breathing,

artery torsion, etc.) is reduced in two stages. First, we discard motionless pixels and, then we select pixels presenting a clear cardiac periodic motion. Motionless pixels are discarded by considering those points with LM cardiac amplitude over a specified range for all LM cardiac amplitudes [10]. The amplitude filter removes those LM signals in which cardiac amplitude was biased from the true one. The remaining LM signals, after motionless pixels removal, should present a periodic profile. However, other dynamic phenomena, such as breathing, morphological changes along the sequence, as well as irregularities in heart beat are prone to reduce the cardiac amplitude and, thus, might distort the ideal discrete profile. Such signals do not properly reflect cardiac motion and should be excluded. The more irregular the profile is, the more spread the frequencies are. We remove such signals by means of an optical filtering [11] centered at the principal harmonic  $\omega_c$ . Optical filtering is a technique widely used in electron crystallography to discard harmonics corrupted with noise. Optical filtering selects only those harmonics presenting a prominent peak. The degree of peak of the harmonic is given by the normalized difference between the amplitude achieved at the harmonic and an average of amplitudes in a neighbourhood  $I_{wc}$  centered at the principal harmonic:

$$OF(\omega_c) = \frac{|\widehat{LM}(\omega_c)|}{S} - \frac{1}{N \times S} \sum_{x \in I_{\omega_c} \setminus \omega_c} |\widehat{LM}(x)|, \quad (1)$$

where  $S = \sum_{x \in I_{\omega_c}} |\widehat{LM}(x)|$  and  $N$  is the number of harmonics in  $I_{wc}$ . Only LM signals with OF above

a given threshold [12] contribute to the average defining the signal reflecting cardiac motion.

### 3.6 Gaussian Filtering

Even in healthy subjects, cardiac frequency does not remain constant along the sequence, which introduces (among other phenomena) irregularities in the cardiac motion profile. Such irregularities distort the cardiac signal and corrupt the location of local extrema in the signal reflecting cardiac motion. It follows that the signal should be filtered. Following the literature, we filter the cardiac profile with a band-pass filter. We consider two families of band-pass filters centered at  $\omega_c$ : Butterworth (B) [6] and Gaussian-based (G) [7]. Filters are given in Fourier domain by the formulae:

$$B(\omega) = B_n(\omega) = \frac{1}{\sqrt{1 + \left(\frac{|\omega - \omega_c|}{0.8\Delta\omega}\right)^{2n}}}, \quad \text{for } \Delta\omega = h\omega_c,$$

$$G(\omega) = G_\delta(\omega) = \frac{1}{\sigma\sqrt{2\pi}} e^{-\frac{(|\omega - \omega_c|)^2}{(2\sigma)^2}}, \quad \text{for } \sigma = \delta\omega_c.$$

For Butterworth,  $n$  is related to the filter decay and  $\delta$  is proportional to its support. Meanwhile for Gaussian based, the decay cannot be handled and only its support may be tuned by its deviation,  $\sigma$ .

### 3.7 Thresholding

Image Thresholding is a simple, yet effective, way of partitioning an image into a foreground and background. This image analysis technique is a type of image segmentation that isolates objects by converting greyscale images into binary images. Segmentation involves separating an image into regions corresponding to objects. We usually try to segment regions by identifying common properties. Or, similarly, we identify contours by identifying differences between regions (edges). The simplest property that pixels in a region can share is intensity. So, a natural way to segment such regions is through thresholding, the separation of light and dark regions. Thresholding creates binary images from grey-level ones by turning all pixels below some threshold to zero and all pixels about that threshold to one. Hence in this module, the intensity values in the range 60 to 100 are considered as normal and intensity values below 60 and above 100 are considered to be abnormal.

### 3.8 Heart Condition Check

In this module we need to check the condition of the heart by determining the heart rate with the help of X & Y coordinate values in the image. Heart rate, or heart pulse, is the speed of the heartbeat measured by the number of poundings of the heart per unit of time typically beats per minute (bpm). The heart rate can vary according to the body's physical needs, including the need to absorb oxygen and excrete carbon dioxide. Activities that can provoke change include physical exercise, sleep, anxiety, stress, illness and drugs. The normal heart rate of a resting adult ranges from 60–80 bpm. Bradycardia is a slow heart rate, defined as below 60 bpm. Tachycardia is a fast heart rate, defined as above 100 bpm at rest. When the heart is not beating in a normal range specified, this is referred to as an arrhythmia, which refers to the abnormal condition of the heart. Hence based on the heart rate we declare condition of the heart.

### 3.9 Report Generation and Mail Facility

In this module we need to enter the credentials like name, age and mail-id, using these credentials the report is generated which consists of Name, age, heart rate and condition of the heart. Later the report is attached to the mail and sent to the mail address specified earlier.

## IV. RESULTS AND DISCUSSION

### HOME PAGE:

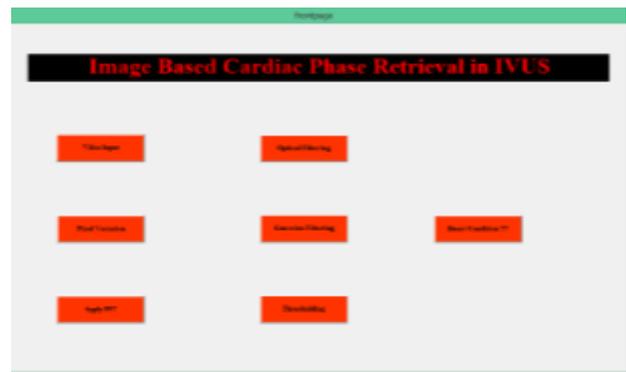


Fig 3. This is the GUI of the proposed system that display links to each operations such as input video, pixel variation, optical filtering, Gaussian filtering, thresholding and heart condition check. When we click on the button, new page will be opened in which the respective operation is carried out

### DATA BASE:



Fig 4. This Window displays the database of IVUS video samples where, we need to select one of the video samples for further processing in the next module.

### LOCAL MEAN EVOLUTION (PIXEL VARIATION):

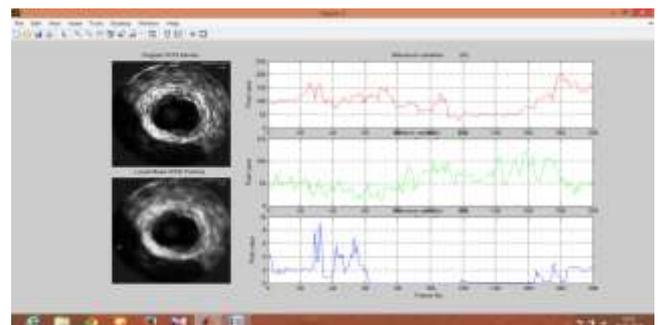


Fig 5. Local mean evolution of different pixels. On the left, the original intravascular ultrasound sequence (top image) and the one corresponding to local mean (bottom image). On the right, the graphics of the evolution of three pixels, a pixel at a salient feature (top), a pixel at lumen-tissue transition (middle), and a motionless pixel (bottom).

**APPLYING FFT:**

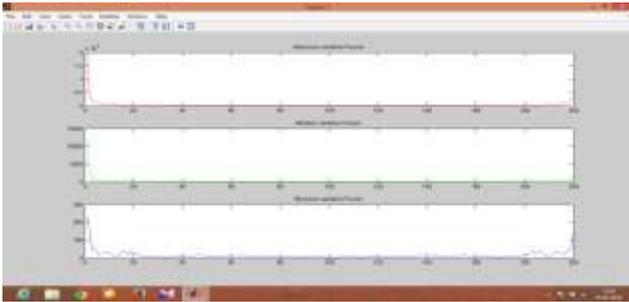


Fig 6. Results showing image of Graph after applying Fast Fourier Transform to the input video sample.

**OPTICAL FILTERING:**

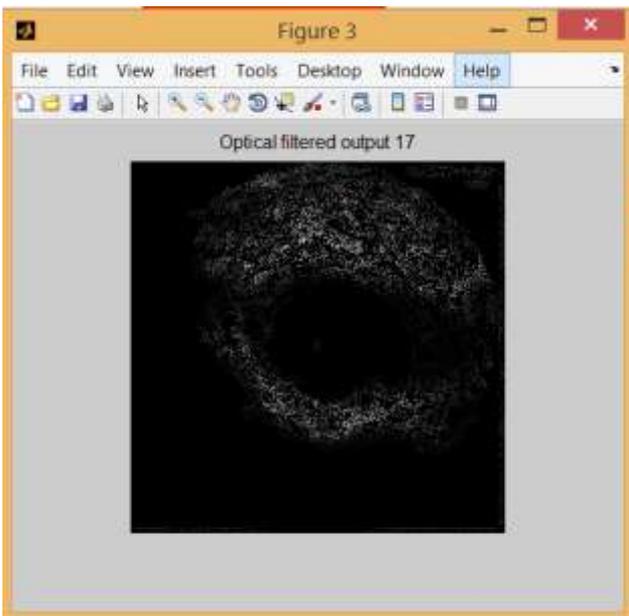


Fig 7. Results showing image of video sample after applying optical filtering.

**GAUSSIAN FILTERING:**

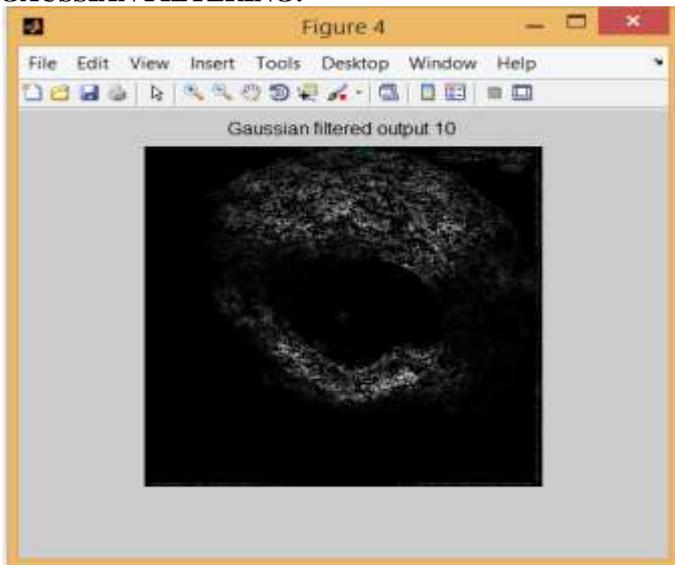


Fig 8. Results showing image of video sample after applying Gaussian filtering.

**THRESHOLDING:**

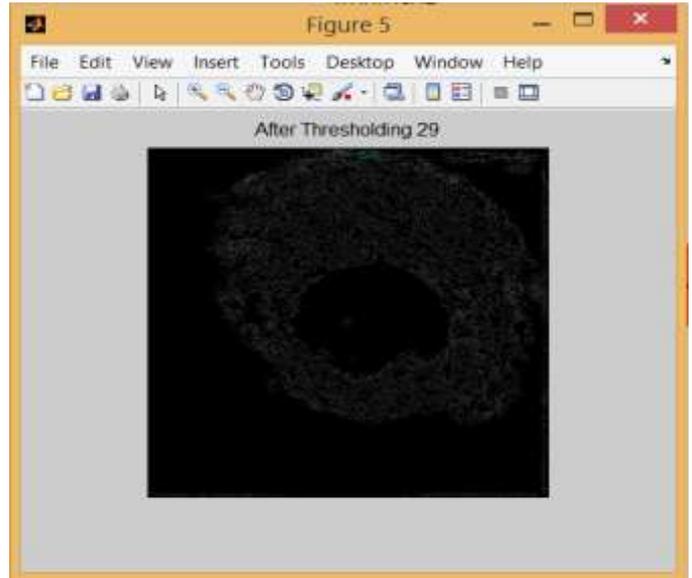


Fig 9. Results showing image of video sample after applying thresholding.

**HEART CONDITION CHECK:**



Fig 10. Results showing the GUI of the heart check condition.

**NORMAL CONDITION:**



Fig 11. Results showing image of Cardiac Phase retrieval as a Normal Condition.

**ABNORMAL CONDITION:**

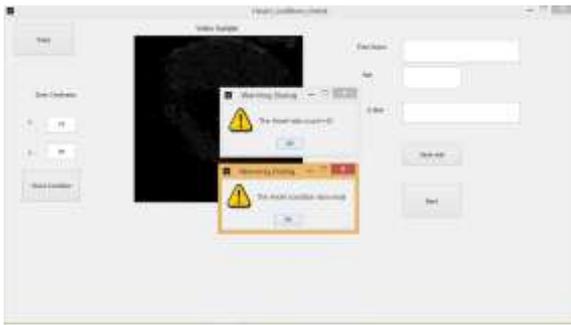


Fig 12. Results showing image of Cardiac Phase retrieval as a Abnormal Condition.

**SENDING REPORT:**

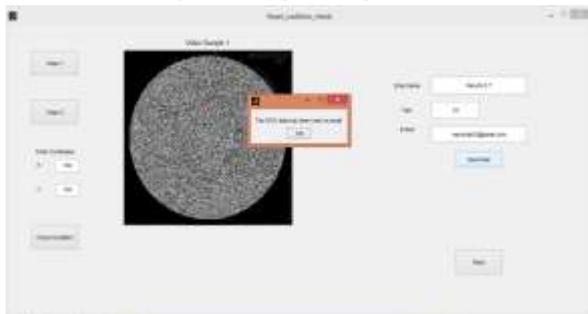


Fig 13. Result showing the image of sending IVUS report to the patient through mail.

**INBOX VIEW:**



Fig 14. Result showing the image of Inbox when we receive the mail.

**REPORT FOR NORMAL CONDITION:**



Fig 15. Result showing the image of IVUS report received for Normal Condition.

**REPORT FOR ABNORMAL CONDITION:**



Fig 16. Result showing the image of IVUS report received for Abnormal Condition.

**GRAPHICAL REPRESENTATION:**

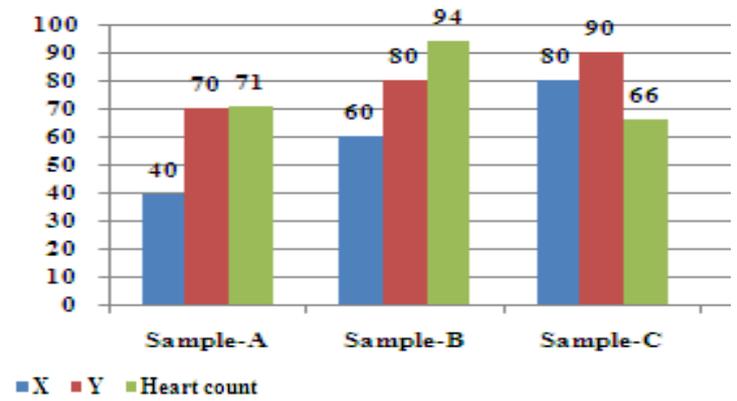


Fig 17. Result showing the image of output analysis for normal video, Here we have considered 3 video samples whose output values are in the range between 60 to 100, which is specified as normal, hence the resulted output is of 100% accuracy.

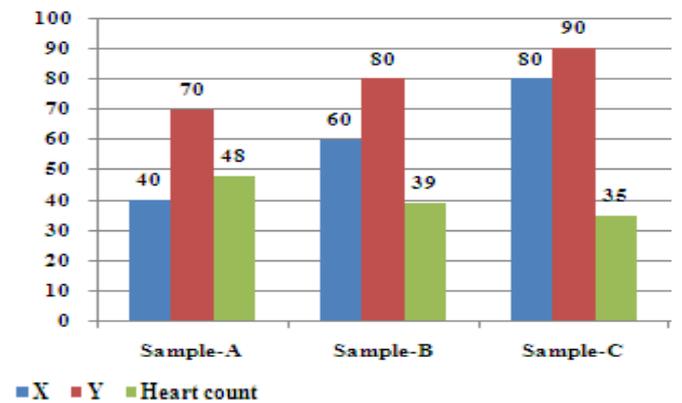


Fig 18. Result showing the image of output analysis for abnormal video, Here the 3 video samples are tested, whose output values are below 60 and the range below 60 is

specified as abnormal, hence the conducted experiment is of 100% accuracy.

## 5. LIMITATIONS

A main limitation of image-based gating methods is that they require an apparent cardiac motion along IVUS sequences. Consequently, accuracy of cardiac phase retrieval is prone to decrease at segments on infarcted hearts presenting a drastically reduced cardiac motion. Given that pathological groups have not been distinguished in our experiments, this might be a main source of error, with an impact on performance that has not been identified. Another source of error in current image-based methods is that they rely on changes observed in sequence intensity. We consider that the quality of IVUS images naturally bounds accuracy of intensity-based methods. It follows that current approaches (including ours) are unable to produce samplings synchronized to the shortest cardiac phases (is volumetric contraction and relaxation). Both phases last between 30 and 70 ms [13], which would imply an average error between  $\pm 1$  and  $\pm 2$  frames. We are currently exploring alternative quantities to image intensity evolution based on vessel dynamics. A main limitation of our validation protocol is the manual sampling of sequences, because it is prone (because of the quality of longitudinal cuts) to present a high inter- and intra- observer variability. This might introduce a source of variability in statistics and it is a main drawback for fine tuning of the filter profile. An on-going work is gathering a database with ECG signals large enough for a better design of the filter profile.

## 6. CONCLUSION & FUTURESPECTIVE

In this paper, we have presented a cardiac phase retrieval system that identifies heart rate and classifies the heart into normal or abnormal category. The system is general enough to be capable of identifying and classifying several factors related to heart while requiring minimum amount of data. Exploring local mean evolution is a fast way to extract cardiac signal from IVUS sequences. We used optical and Gaussian filtering for the removal of unwanted signals. The benefit of using this system is that, it is efficient than the existing systems specified in the literature. The proposed work classifies the heart as normal or abnormal based on the range specified. Simultaneously the IVUS report is generated which consists of the parameters regarding heart condition, which is mailed to the concerned. The accuracy compares to existing approaches but significantly reduces computational complexity. However, since cardiac phase can be strongly affected by artery lesions and other cardiac factors, a study in depth of the filter parameters would check the robustness of the system to a variety of subjects. This

fact implies that a larger experimental setting would contribute to solid results.

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