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Time and Frequency Domains Analyses of Atrial Fibrillation Activation Rate: The Optical Mapping Reference

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Abstract

Background—Time- and frequency-domain estimates of activation rate have been proposed to guide atrial fibrillation (AF) ablation in patients but their electrophysiologic correlates are unclear.

Objective—To study the relative correlation of average electrical cycle length (CL) and dominant frequency (DF) during AF with reference optical mapping measures.

Methods—Eight sheep hearts were Langendorff-perfused and superfused with oxygenated Tyrode's solution inside a tank representing the human thorax. Optical mapping (DI-4-ANEPPS) of 4×4 cm² in the left atrium was performed at 0.5 mm/pixel and 600 fps. A 20-pole catheter was placed in the optical field of view to acquire 1.2 kHz unipolar recordings by the EnSite NavX™ System (ENS) optimized for CL and DF calculation. During AF, 5-sec long simultaneous optical and electrical signals were analyzed for CL and DF.

Results—During pacing, DF measurements had less false results than CL (6.6–2.5% vs. 21.5–4.4% depending on filtering, $p < 0.001$). During AF in regions showing periodic waves on both sides of the catheter optical 1000/CL vs. DF correlation showed 95% confidence identity and was better than unipolar measurements in the ENS (Adjusted R²: 0.58879 vs. 0.12902; $p < 10^{-6}$). DFs of unipolar signals correlated better than CLs with DFs of optical signals. Similarly, bipolar DF correlation with optical DF was not different from identity ($p > 0.157$), but the bipolar CL showed smaller identity with the optical CL ($p < 0.0004$).

Conclusion—DF values of unipolar and bipolar signals correlate with those of optical signals better than CL values for the respective signals.

Keywords

Atrial fibrillation; Complex fractionated electrogram; Cycle length; Dominant frequency; Ablation

INTRODUCTION

Analyses of intra-atrial electrograms in the time domain^{1,2} and frequency^{3,4} domain can be used to guide ablation procedures and increase their efficacy in patients with either paroxysmal or persistent AF. Two distinct approaches are typically used; one that highlights electrogram irregularity,⁵ and the other that highlights activation frequency.⁶ Both approaches point to the importance of short cycle lengths and high frequency as measures of high number of activations per unit time (activation rate) and maintenance of AF. Thus the EnSite NavX™ System (ENS; St. Jude Medical) calculates two parameters for each signal:

The first one is the time domain mean Complex Fractionated Electrogram (CFE-mean) parameter, which measures an average interbeat interval (CL) over a predetermined recording period. The second is the Dominant Frequency (DF) parameter which is the frequency with the highest power in the power spectrum representing the average excitation rate.⁶ Concurrently, increasing numbers of studies have demonstrated a mismatch between these two measures of the activation rate.^{7–11}

In experiments using animal models of AF, optical mapping of the cardiac action potential offers a unique approach for broad-view characterization of complex propagation patterns with high spatiotemporal resolution. Importantly, studies based on both optical and bipolar signals have found that the area with the highest activation rate as quantified by the DFs correspond to the posterior left atrial wall, where AF drivers were identified.^{12–14} Thus, this study aims at enhancing the ability to interpret average CL and DF, which are used by the ENS and comparable devices as measures of local activation rate during AF, by correlating them with similar measures obtained in optical mapping. Based on a recent study,¹⁵ we hypothesized that DF will exhibit a higher correlation between the electrical and the optical modalities when compared with the CL correlation. To test this hypothesis we used Langendorff-perfused sheep hearts and simultaneous ENS and optical recordings. Our analyses demonstrate that for both unipolar and bipolar signals, the DF parameter correlates better than the CL with the optical data.

METHODS

Animals and experimental system

All animal experiments were carried out in accordance to the Guidelines for the Care and Use of Laboratory Animals of the National Institute of Health. Eight sheep hearts were Langendorff-perfused inside a transparent Plexiglas tank (see Figure 1, panel A) representing the human thorax. A CCD camera and the ENS systems recorded simultaneously fluorescence and unipolar voltage, respectively, during pacing and sustained AF (Figure 1, panels B and C; see also OS). Panel D of Figure 1 shows a sample of 20 simultaneously ENS recorded traces during sustained AF. The bottom trace is a synchronizing camera signal indicating initiation of optical recording (red arrow).

Analysis of the electrical recordings

Four different filtering ENS modes were applied to the unipolar recordings: 1) No filtering (labeled as None); and band-passes of 2) 12–300 Hz (L300); 3) 12–150 Hz (L150); and 4) 12–50 Hz (L50).

On the Diagnostic Landmarking Map module of the ENS the CFE-mean-parameter represents the average time interval between sequential activations. The same module also calculates DF for the same signals representing the maximal power frequency. The settings of the module for the CFE-mean and DF calculation of the unipolar recordings were adjusted by optimizing the detection of the pacing rate at the maximal number of electrodes for each experiment(see OS).

Bipolar signals were generated offline by subtraction of exported unipolar ENS recordings from adjacent electrodes. CL and DF for the bipolars were determined with pre-existing Matlab algorithms (see OS).^{3,4,15}

Statistics

For each modality data set analyses were performed blind to comparisons. Linear regression and correlation analyses were performed (Origin 8.1; OriginLab Corporation, Northampton,

MA) and displayed on relevant graphs as the best fit (red lines), 95% confidence boundaries (blue lines), 95% predictable scatter boundaries (purple lines) and an identity reference line ($y=x$, black line). R^2 are reported following adjustment (AR^2). Correlation coefficients were compared following Fisher's z -test and slopes were compared using Student's t -test (see Online Supplement, OS). $P<0.05$ was considered significant.

RESULTS

Pacing and AF analyses using the ENS

We compared the CFE-mean and DF values calculated by the ENS for 20 unipolars from a catheter secured as seen in Figure 1C. The histograms in Panel A of Figure 2 show the distribution of 1000/CL and DFs in the ENS as determined for a total of 158 recordings from 8 animals during pacing at 200 ms cycle length under the L50 and L150 filtering modes. Visual examination of the 4 histograms reveals that most of the electrodes reproduced the expected result of 5 Hz, but they were skewed differently for CL versus DF; those based on CL had false detections at $1000/CL < 5$ Hz (i.e., CLs detected were >200 ms) and those based on DF had false detections at $DF > 5$ Hz. Quantification of the histograms for the L150 filter yielded 44/316 recordings with false 1000/CLs and DFs detections; that number reduced to 11/316 when L50 filtering was used ($p<0.001$ Fisher's exact test). Overall, DF measurements had less mismatches with the pacing cycle length than 1000/CL (6.6 and 2.5 % vs. 21.5 and 4.4 % in L150 and L50 filtering, respectively. $p<0.001$, Fisher's exact test).

Once the settings for CL detection were adjusted for optimal performance during pacing they remained fixed for the analysis of AF. According to theory, 1000/CL and DF should represent the average activation rate during the period analyzed and therefore should correlate to each other with an identity line of slope=1 and no offset. Yet, the correlation that was found between these two parameters during AF was very low and the linear trend slope was far from 1. Panel B in Figure 2 shows plots of 1000/CL vs DF for 158 points determined during 8 episodes of AF with settings of L50 (top) and L150 (bottom) filter. A linear best fit shows a trend of $y=9.54+0.193x$ and $AR^2=0.075$ for the L50 and $y=5.989+0.189x$ with $AR^2=0.049$ for L150. Based on their respective fitted slope's standard error of 0.05219 and 0.06021 we determined that the slopes of 1000/CL vs. DF are different from 1 ($p<0.0001$). The $<<1$ slope of the relationship could be explained by the oppositely skewed histograms of the detected CLs and DFs in panel A. To better characterize such low correlation values we proceeded to compare the analysis of electrical recordings with that of optical recordings that would serve here as a reference for the action potential activation rate.

Wave-by-wave optical-electric comparison

The CCD camera system cannot give simultaneous information at the exact same location where the electrical signals are recorded because the tissue is masked by the circular electrode. To increase reliability of the correlation between optical and electrical data we performed those comparisons only in five 5 second-long episodes showing AF waves that traveled repeatedly on the two opposing sides of the catheter with a relatively fixed interbeat interval for 4 or more cycles (i.e., waves exhibiting spatiotemporal periodicity, STP). These included waves that traveled uninterruptedly either across or in parallel with the catheter to allow interpolation of the optical data with the electrical signals from the catheter that masked the tissue underneath. Panel A of Figure 3 shows an example of such STP waves: Five (out of eight observed) sequential impulses are shown to cross (white arrows) the circular catheter (gray outline) at about 6–7 o'clock. Waves traveling on the two sides of the catheter, but parallel to it, were also identified but not shown here. In most cases, the STPs

during the AF episode did not last for the entire 5-second duration of the recording, but rather they altered their patterns.

Superimposed on the leftmost activation map of Figure 3A are two markers indicating locations selected for pixel signals for the comparison with the electrical recordings. Panel B of Figure 3 shows 260 msec-long samples out of 5-sec-long time-series and their corresponding power spectra obtained simultaneously from optical and electrical recordings in a pair of flanking pixels and a pair of electrodes between them. The top and bottom traces are signals from the pixels marked on the activation map. They show the typical oscillatory feature of activation during fibrillation as detected by fluorescence mapping. These optical traces flank electrical ENS recordings from the catheter electrodes between the two pixels. All electrical traces correspond to the same location and timing, but are grouped for the 4 different processing routines employed: unprocessed signal (None) and the L300, L150 and L50 filtering modes.

Superimposed on the optical and electrical recordings are the respective detected activation times. For the optical traces the 50% amplitude level was marked down as the activation (red circles with green and purple vertical lines). For the ENS-based unipolars and bipolars the time of activation was determined here as the minimal time-derivative (red and blue triangles) and maximal deflection (black triangles), respectively. Comparing the 4 main unipolar and bipolar deflections in Figure 3B, one notices a general qualitative correspondence to the fluorescence deflections. However, differences become evident when considering the activation times: only the fourth electrical activation coincides with the optical activation (green and purple vertical lines, except L50) while the other waves appear more delayed. Note also that as more aggressive filtering is applied (i.e., from None, to L300, L150 and finally L50) the activation times are progressively more delayed relative to the pixels from about 10–20 ms for the None, L300 and L150 signals to about 20–30 ms for the L50 signal, suggesting that processing with the L50 filtering mode introduces phase shifts into the analyzed signals. Overall, in 81% of waves analyzed in 4 different animals during 5-sec long episodes of AF, the L50 filtering setting results in delayed activation time detection by the bipolars relative to the corresponding pixels. The best coincidence between maximal voltage activation times of bipolars and optical signals was obtained with the L300 filtering setting. However only about 35% of the bipolar waves were fully concordant with the flanking pixels. In about 37% of waves the bipolars activated earlier and in 28% they activated later than the flanking pixels, often within the same recording.

The automatic 5-sec average CLs for the signals exemplified in Figure 3B ranged as follows: optical signals, 60–71 ms; unipolars, 55–65 ms; bipolars, 60–76 ms. The corresponding power spectra, on the other hand, showed an invariable DF of 17 Hz (59 ms) for all the signals. Manual counting by two investigators of the number of activations for the 5-sec optical signals yielded 86–91 activations, translating into an activation rate of 17.5–18.5 Hz (CL of 57–54 ms, respectively; see OS and Figure OS1). The reason for the variability in the relative timing of activations and rates is unknown; it may be related to complex spatiotemporal patterns of activation in the small area comprising the pixels and the two recording unipolars¹⁶ and/or different noise handling. In any event, the same variability may underlie discrepancies between electrical versus optical and time versus frequency methods shown further below and becomes important when using one type of recordings and analysis as a reference.

Correlating optical and electrical CL vs DF

Prior to comparing the time- and frequency-domain analyses of electrical versus optical signals we wish to establish the baseline properties for these domains in their respective recording modality. Figure 4 shows point by point comparisons and correlations between

1000/CL and DF values for 20 single pixel recordings (panel A, see also Figure OS1) and 30 ENS unipolars (panel B) in the same area and episodes. Most of the points do not fall exactly on the desired identity line for either the optical or the electrical modality. The AR^2 for the optical correlation is nevertheless significantly larger than the electrical correlation (0.58879 and 0.12902, respectively. $p < 10^{-6}$). While for the optical relationship (panel A), the line $y=x$ lies within the 95% confidence level, this is not the case for the unipolar data relationship (panel B). In fact, although the AR^2 of 0.12902 for the electrodes here is higher than the AR^2 of 0.075 in Figure 2 for the indiscriminate selection of electrode recordings, the correlation is nevertheless not significant ($p=0.2845$).

CL and DF in unipolars vs. optical signals

At this point it is evident that the correlation between time- and frequency-domain measurements is different for the electrical versus optical data we analyzed. To further understand that discrepancy we proceeded to study the performance of the time- and frequency-domain analyses separately for the electrical and optical modalities. In Figure 5 we present our analysis for the same areas as in Figure 4. The unipolar signals included those subjected to L50 and L150 filtering modes to test for the possible effect of phase shifts. Panel A shows the time-domain analysis comparing the CFE-mean of the activity in the ENS unipolars and the optical signals. As can be appreciated, the line of identity $y=x$ falls mostly outside the 95% confidence interval for both the L50 and L150 modes with respective AR^2 values of -0.02621 and 0.01857 , leading to the conclusion that the average CL of the unipolars does not correlate with the average CL of the optical data. In panel B we show the frequency-domain analysis. In contrast to the CL correlations, the 95% confidence interval of the unipolar-optical DF correlation includes the identity relationship $y=x$ for both L50 and L150 filtering modes (see OS Table 1 for best fit parameters). The DF AR^2 value of 0.67484 for L150 is higher than the value of 0.31067 for L50 ($p < 10^{-6}$) and both DF AR^2 values are higher than those of the CL ($p < 10^{-6}$). See below for a comparison of the slope values of the DF and CL correlations.

CL and DF in bipolar vs. optical signals

In Figure 6 we analyze the CL and DF correlations using bipolar signals. The analysis of bipolar signals was performed using custom-made routines that were not significantly different from the unipolars ENS routines ($p=0.185$ Fisher's exact test, see OS). During AF, as with the unipolar signals, AR^2 shows a trend for higher values in the correlations of DF versus CL (see OS Table 2). However this trend is not statistically significant. In panel A of Figure 6 the identity line falls largely outside the 95% confidence interval for the CL correlation, which is also partially true in panel B for the DF correlations. The graphs in Figure 6 nevertheless show that the main difference between the CL and DF correlations is in the level of the dispersion of the bipolar parameters versus the dispersion of the optical parameters.

Dispersions of CL and DF in electrical vs. optical signals

In a recent study, Atienza et al⁴ demonstrated that abolishing by ablation pre-existing AF DF gradients predicts long-term freedom of AF in humans, consistent with earlier studies using optical mapping in sheep hearts.¹³ Thus it is important to assess whether the dispersion of the electrical data correlates with the optical data. In Figure 7 we compared the slope values of the best fitted linear regressions for the unipolar and bipolar signals. In panel A, the values of the slopes for the 4 different unipolar vs. optical correlation graphs (see Figure 5) reveal a large sensitivity of the unipolar signals to the L50 vs. L150 filtering mode. For the L50 the slope was negative; for the L150 the slope has a large standard error which makes it not significantly different from either 1 or zero ($p > 0.1003$), making the CFE-mean correlation impractical. On the other hand, the slopes of the ENS DF regression lines for

both filtering modes (white bars), are not different from the desired 1 ($p > 0.2644$). In Panel B, the linear regression slopes were compared similarly for the correlations of the bipolar signals (see Figure 6). Regardless of the filtering mode, the slopes of the bipolar DF vs. optical DF (white bars) were not different from 1 ($p > 0.57$) and the bipolar CL slopes were significantly smaller than 1 ($p < 0.0004$). Altogether, our comparisons of slopes demonstrate that the DF dispersion determined for both unipolar and bipolar signals follows the dispersion of the optical signals closer than the CL dispersion.

DISCUSSION

In this study we compare directly for the first time data obtained using a clinical electro-anatomical mapping system with optical mapping data, and correlate time- and frequency-domain methods of quantifying local activation rate. Mapping the electrical activity of an isolated ovine heart during pacing and AF simultaneously with the ENS and a fluorescence imaging system demonstrates that: 1) Neither optical nor electrical recordings show a CL vs. DF point-by-point match, 2) In the optical modality, however, the correlation between the time- and frequency-domain parameters is not significantly different from $1000/CL = DF$ (i.e., an identity relationship). 3) Both ENS unipolar and bipolar DFs show better correlation with optical DFs than the CLs. In sum, our study demonstrates a more robust correlation of the local activation rate in electrical vs. optical signals by DF quantification than by the average CL quantification.

Optical mapping as a reference to characterize activation during fibrillation

The impact of the optical mapping approach in studying AF may rely on what distinguishes it from the electrical recordings:^{17–19} 1) High-resolution over areas larger than a typical wavelength of the propagating impulses during AF. 2) The fluorescence signal is more directly linked to the membrane potential than the electrode signal, which measures extracellular potential. 3) The optical signal is less dependent on the direction of impulse propagation compared with bipolar or large unipolar electrodes. For all those reasons, spatiotemporal optical data during fibrillation is readily interpretable with minimal processing compared to electrode recordings and provides solid information on impulse propagation and the number of activations per unit time.

Time- versus frequency-domain determination of activation rate

Previous studies presented evidence that the frequency-domain derived DF parameter can be used to detect activation rate despite beat-by-beat variations in the activation amplitude and CL.^{11,20–22} However, prior to the present study no comparisons had been presented between DF and the independent time-domain methods. Here we correlate independent time and frequency domain methods of estimating the rate of local activation during fibrillation using electrical and optical approaches. The correlation between the $1000/CL$ and DF in the optical modality shows 95% confidence, but such a correlation does not exist for $1000/CFE$ -mean of unipolars analyzed in the ENS (Figure 4). One obvious possibility for such a discrepancy is that both CL and DF are prone to uncorrelated variability (i.e., false detections). Since the time-domain versus frequency-domain detection of activation rate in the ENS system is uncorrelated, then one should ask whether the user should adopt the system's time or its frequency domain method for the estimation of the activation rate. In simulations it has been demonstrated that the greater the variation in beat-by-beat amplitude and interbeat intervals, the less likely the DF will match the pre-assigned mean activation rate.^{20,21} However, Grzeda et al¹⁵ demonstrated numerically that the reliance of the time domain method on each activation time renders that approach more sensitive to signal morphology, noise and selection criteria when compared with the frequency domain method.

This is consistent with our results in Figures 5–7 showing that DF detection for either unipolar or bipolar signals correlates better with the optical signals than the CL detection.

Limitations

The detection algorithms and settings employed in this study are those in the ENS, as well as custom made Matlab routines which may differ from other clinical systems. In particular, the ENS settings were adjusted to optimize unipolar matching with paced activity and used also during AF, where signals may differ in their properties. In the future, a better adjustment approach should be considered, but the present conclusions are supported by the equivalence of the approaches applied to both time- and frequency-domain methods with unbiased presence of both under- and over-sensing and by similar conclusions for exported data. We also note that the use of a 12 Hz high-pass and 50/150 Hz low-pass filtering as employed in this study is not common in clinical setting and may require further adjustments.

Clinical implications and conclusion

Studies have suggested that determining the distribution of the complex fractionated atrial electrograms (CFAEs)⁵ through CFE²³ and activation rates through DFs^{3,4} across the atria in patients with either paroxysmal or persistent AF can guide ablation procedures and predict long term freedom of arrhythmia. Unfortunately, dye toxicity and optical issues precludes the use of optical mapping in patients. Nevertheless here we show that electrical estimation of activation rate via DF analysis correlates with optical mapping data and provided a basic science rationale for the clinical observation.⁴ In particular, we submit that when using the ENS for AF analysis in patients, the DF option will be preferable over, or should be used in conjunction with, the CFE-mean option for more physiologically relevant determination of the spatial distribution of the activation rates.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Disclosures

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ABBREVIATIONS

AF	Atrial fibrillation
AR	Adjusted regression coefficient
CFE	Complex fractionated electrograms
CL	Cycle length
DF	Dominant frequency
ENS	EnSite NavX™ System

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Figure 1.

The electro-optical experimental setting. A) A front view of the isolated Langendorff-perfused sheep heart inside the 30-L transparent Plexiglas tank. A CCD camera is seen on the right side B) A left view of the isolated heart through the transparent Plexiglas wall. The catheter is seen to be in contact with the LA epicardium (blue arrow). Two sample electric-field generating electrodes are marked on the tank wall (red arrows). C) A 20-electrode circular catheter is seen through the CCD camera to be stitched onto the LA free wall/appendage (LAFW/LAA; Ant, anterior; Post, posterior). D) A sample snapshot of the ENS screen showing 20 unipolars recorded during AF. The bottom trace is a digital input with a step indicating the instant of the beginning of a movie recording (red arrow).

A (During 5 Hz pacing) **B (During AF)** **Figure 2.**

ENS Diagnostic Landmarking module analysis for CFE-mean (average CL) and DF from 158 unipolar recordings using the 20-pole catheter (N=8, 5-second long episode per heart). A) Histograms show the distributions of the CFE-mean and the DF for the L50 (top) and L150 (bottom) filtering modes during pacing at 200 ms (5 Hz). The settings in the Diagnostic Landmarking module were optimized to maximize the number of pacing rate detections by the CFE-mean and DF. B) Cumulative correlation of the ENS 1000/CFE-mean (1000/CL) and DF during 8 episodes of AF for the L50 (top) and L150 (bottom) filtering modes.

Figure 3.

A) Activation maps showing sequential waves propagating in the direction of the white arrow and crossing the circular catheter (superimposed gray shade) near electrodes 11 and 12 in this animal. Sites 1 and 2 in the left-most panel indicate pixels used for optical data analysis and correlation with electrodes signals. B) Sample comparison between simultaneous nearby optical (top and bottom) and electrical (middle; u, unipolars; b, bipolars) time and frequency analysis. Left: sample 260 msec episode showing STP during AF. Red circles: Activation times of optical signals. Red and blue triangles: Activation times of the unipolars. Black triangles: Activation times of the corresponding bipolars. Green line: Activation times of pixel 1 (top). Purple line: Activation time of pixel 2 (bottom). Right: corresponding power spectra of 5-second long signals including the episodes shown on left. Circles and triangles: Dominant frequency peaks (blue triangles are overlapped by red triangles).

Figure 4. Correlations between 1000/CL and DF values for 20 single pixel recordings (panel A, N=5) and 30 unipolars recorded by the ENS and analyzed by the CFE-mean (panel B, N=5). The data points in the two panels were obtained in the same atrial region and during same 6 AF episodes. See Methods/Statistics for line colors legend.

Figure 5. Correlations between unipolar (ENS) and optical signals (N=5, during 6 movies). A) Correlation of ENS CFE-mean and optical CL for L50 (top, n=40) and L150 (bottom, n=38). B) Correlation of ENS DF and optical DF for L50 (top, n=28) and L150 (bottom, n=28). See Methods/Statistics for line colors legend.

Figure 6. Correlations between bipolar and optical signals. A) Correlation of CL for L50 (top, n=24) and L150 (bottom, n=24). B) Correlation of DF for L50 (top, n=32) and L150 (bottom, n=32). See Methods/Statistics for line color legends.

Figure 7.

Comparisons of dispersions of the CLs and DFs between electrical and optical modalities.

A) Values of slopes for the 4 different correlation graphs shown in Figure 5 for unipolars (ENS CFE-mean and DF) vs. optical data. B) Values of slopes for the 4 different correlation graphs shown in Figure 6 for bipolars (CL and DF) vs. optical data. Horizontal red line: Desired identity slope value of 1. X-axis labels refer to either CL (gray bars) or DF (white bars) with the respective filtering mode (L50 or L150).