

Original Article

Comparison of complex fractionated atrial electrograms between the left atrium and pulmonary veins in development of persistent atrial fibrillation in goats

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Abstract: Background: The role of complex fractionated atrial electrograms (CFAEs) in the maintenance of atrial fibrillation (AF) is controversial. The main problem is the underlying mechanism of CFAEs is not clear. Objective: This study aimed to compare CFAEs between the left atrium (LA) and pulmonary veins (PVs) dynamically at development of and terminations stage of persistent AF in goats and try to explore the mechanism of CFAEs. Methods: Eight female goats were instrumented with electrodes at the LA and left side PV. Sustained AF (>24 h) was induced in the goat by rapid intermittent left atrial pacing for 9.3 ± 4.6 days. After AF lasted for more than 24 h spontaneously, propafenone was infused intravenously until termination of AF. Characteristics of PV and LA electrograms were analyzed in the development and termination of AF. Results: With prolonged stimulation, the duration of AF prolonged, CFAEs in LA and PVs increased gradually, PVs have more CFAEs than LA all the time. When induced AF lasted for more than 24 h, CFAEs in PVs became sustained approximately ($3.3\% \pm 4.0\%$ vs. $91.8\% \pm 6.7\%$, at onset of AF vs. AF lasted for more than 24 h, $P < 0.05$), and the ratio of CFAEs in PVs was more than that in LA ($91.8\% \pm 6.7\%$ vs. $78.7\% \pm 4.6\%$, $P < 0.05$). Administration of propafenone resulted in a gradual decrease of CFAEs in the LA and PVs ($P < 0.05$). CFAEs disappeared in the LA before cardioversion. Sinus rhythm resumed only when CFAEs in the left superior PV vanished completely. Conclusion: CFAEs are area-specific. CFAEs are increased gradually with the electrical remodeling. CFAEs in PVs may play an important role in the maintenance of AF in this model.

Keywords: Atrial fibrillation, fractionated electrogram, pulmonary vein, left atrium, propafenone, goat model

Introduction

Complex fractionated atrial electrograms (CFAEs) are defined as electrograms with two or more deflections or electrograms with continuous deflection over a 10-second period or as electrograms with mean cycle length ≤ 120 ms over a 10-second period [1]. Mapping and ablation of CFAEs is a novel approach for atrial fibrillation (AF). Nademanee et al. [1]. Initially described CFAEs as an ablation target for AF. They found that ablation of CFAEs led to acute termination of AF in 95% of cases, and this single procedure led to 76% freedom from AF at a 1-year follow-up. Nevertheless, only few studies indicated the adjunctive CFAEs ablation could provide additional benefit for patients with nonparoxysmal AF [1-3]. Most studies show

that no reduction in the rate of recurrent atrial fibrillation when ablation of CFAEs was performed in addition to pulmonary-vein isolation [4-7]. Some scholars argued that the negative results should be attributed to the useless of automatic CFAEs detection algorithms [8]. Also, the ablation strategy of the following studies are different from that of Nademanee. But, the main problem is that the underlying mechanisms of CFAEs and the role of CFAEs in occurrence, development, and termination of AF are unclear.

Experimental studies in canines showed fractionation of pulmonary veins (PVs) electrograms in pacing-induced sustained AF, suggesting that the PVs may play an important role in maintaining AF [9-12]. However, to the best of our knowledge, few studies have focused on the dynamic

The dynamic changes of CFAEs

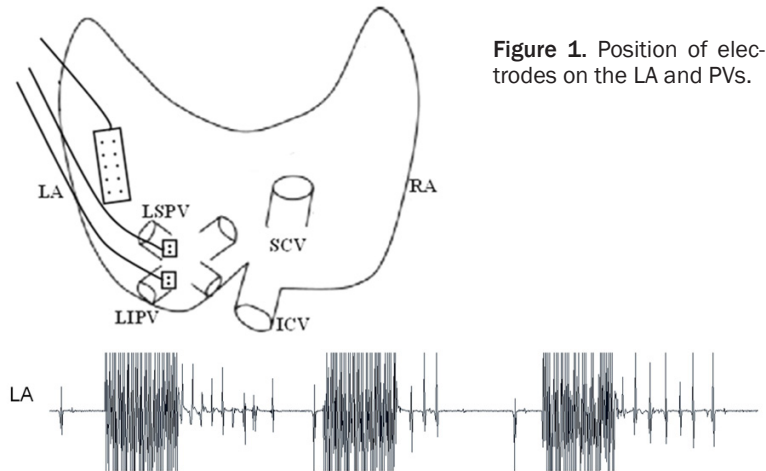


Figure 1. Position of electrodes on the LA and PVs.

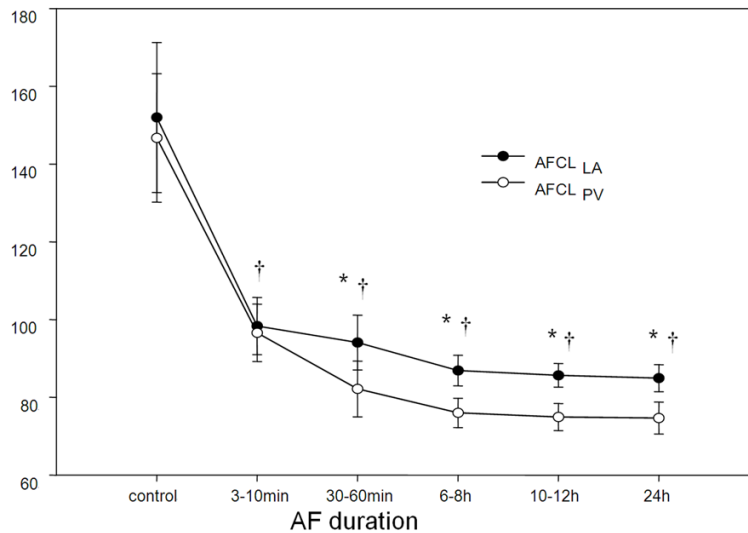


Figure 2. Changes in AFCL within the LA and PV with prolonged atrial fibrillation. AFCL = atrial fibrillation cycle length; $AFCL_{PV}$ = AFCL within the pulmonary vein; $AFCL_{LA}$ = AFCL within the left atrium; $^{\dagger}P < 0.05$ compared with baseline; $^{*}P < 0.05$ compared between $AFCL_{LA}$ and $AFCL_{PV}$.

changes of CFAEs in PVs regarding the occurrence, development, and termination of AF. Additionally, whether development of CFAEs in PVs is a major factor in self-perpetuation of AF is unclear. This study aimed to observe the onset, development, and termination of CFAEs in pacing-induced sustained AF in goats. This study also aimed to compare the CFAEs between the left atrium (LA) and PV during occurrence and termination of AF to determine the role of CFAEs in PVs in persistent AF.

Materials and methods

Eight adult female goats (3.3 ± 1.2 years old), weighing between 28 and 45 kg (37.4 ± 6.3 kg),

were used for this study. All protocols followed US Department of Agriculture and National Institutes of Health guidelines and were approved by the Animal Investigation Committee of the Chinese PLA General Hospital. The goats were anesthetized with 3% isoflurane and a 2:1 mixture of O_2 and N_2O . Left intercostal thoracotomy was performed and the pericardium was opened to expose the heart. A 10×1.2 -cm silicon strip, containing five pairs of silver electrodes (diameter, 1.5 mm; inter-electrode distance, 5 mm), was sutured to the anterior wall of LA silicon patch measuring 1×1 cm, containing one pair of silver electrodes (diameter, 1.5 mm; inter-electrode distance, 5 mm) was sutured to the roots of the left superior pulmonary vein (LSPV) and left inferior pulmonary vein (LIPV) (**Figure 1**). After approximation of the pericardium and closure of the thorax, the electrode leads were tunneled to the neck. Ampicillin (1 g) was administered prophylactically before and after surgery.

Two weeks after surgery, the goats were connected to an external automatic atrial fibrillator. Goats were kept in separate boxes with free access to food and water. A cable from the top of the box was plugged into the connector in the neck of the goat and the atrial electrodes were connected to a multichannel electrophysiological recorder (GY-6328, South China Medical Electric Company, Henan, China). The atria could be stimulated through any pair of the epicardial electrodes. The custom-made pace maker could generate burst pace at a pacing interval of 20 ms for 1 s with a maximum output of 6.0 V, followed by a 2-s period without pacing [13].

After the goats had recovered from surgery and before they were connected to the pace-

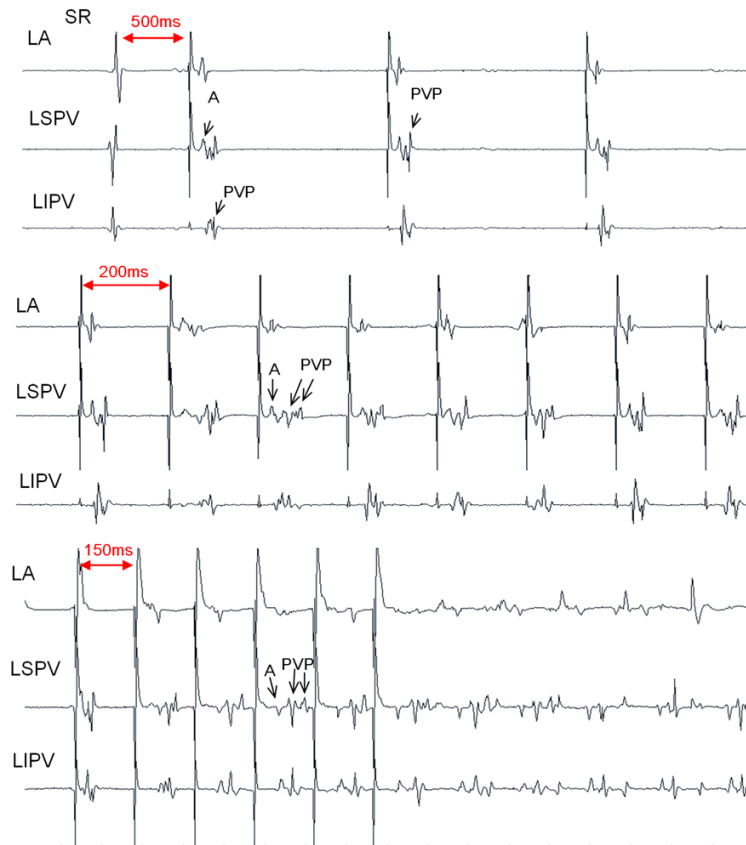


Figure 3. Example of development of fragmentation of a pulmonary vein electrogram during an increase in the left atrial pacing rate. Top panel: pulmonary vein potentials (PVPs) could not be detected in sinus rhythm. PVPs were unmasked by LA pacing with a basic cycle length (BCL) of 500 ms. Middle panel: The BCL was 200 ms and PVPs became fragmented. Bottom panel: the BCL was 150 ms, PVPs were fragmented, and atrial fibrillation was induced. LA = left atrium; LIPV = left inferior pulmonary vein; LSPV = left superior pulmonary vein; SR = sinus rhythm.

maker, the atrial effective period was measured during a wide range of pacing frequencies (S_1S_1 interval, 200-600 ms). A single premature stimulus ($4 \times$ threshold) was interpolated at every eight basic intervals and, starting from well within the refractory period, the S_1S_2 coupling interval was incremented in steps of 2 ms. The shortest S_1S_2 interval resulting in a propagated atrial response was taken as the atrial effective refractory period (AERP).

After the fibrillation pacemaker had been turned on, the pacemaker was disconnected after 6, 12, and 24 h, and every other 24 h, respectively. The duration of AF was recorded until the induced AF lasted for more than 24 h.

The AERP was measured again after 6, 12, and 24 h, and every other 24 h. The duration of induced AF and AF cycle length (AFCL) were

recorded. When the induced AF lasted more than 3 min, the atrial refractory period during AF (RP_{AF}) was measured with the method described by Wijffels et al. [14] and Shan et al. [13]. The stimuli were synchronized with the fibrillation electrograms recorded from the pacing electrode. The RP_{AF} was determined by a single stimulus that was applied after every eight sensed beats through a pair of electrodes on the anterior wall of the LA. Stimulus strength was four times the capture threshold and the duration was 2 ms. The coupling interval was incremented in steps of 2 ms. Each stimulus was repeated 10 times; the shortest coupling interval that captured the atrium more than two of 10 times was taken as the RP_{AF} . The capture criteria were described by Wijffels et al. [14]. The average of 200 consecutive intervals was taken as AFCL. The RP_{AF} was measured every 24 h.

The epicardial atrial electrograms at the LSPV, LIPV, and LA were recorded during sinus

rhythm, after LA stimulation at different pacing intervals (400, 300, 200, and 150 ms), and during AF. All electrograms were classified as single potential, double potential, or fragmented potential [15]. Double potentials were defined as two deflection separated by 15 ms to 50 ms, and the amplitude of the second component had to be at least 25% of the main deflection. Fragmented potentials were defined as three or more deflection separated by more than 15 ms. The ratios of all types of electrograms were counted in the range of more than 300 consecutive activations. The morphology of electrograms in the PVs and LA and the relationship between the ratio of CFAEs and duration of AF were analyzed.

After AF lasted for more than 24 h spontaneously, propafenone was infused by 2 mg/kg

The dynamic changes of CFAEs

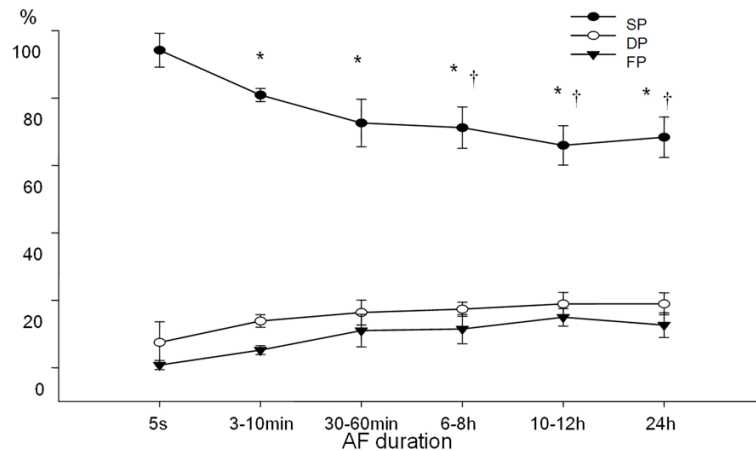


Figure 4. Changes in potentials in the LA with prolonged atrial fibrillation. LA = left atrium, SP = single potential, DP = double potential, FP = fragmented potential. * $P < 0.05$ compared with the occurrence of atrial fibrillation; † $P < 0.05$ compared with atrial fibrillation lasting for 3-10 min.

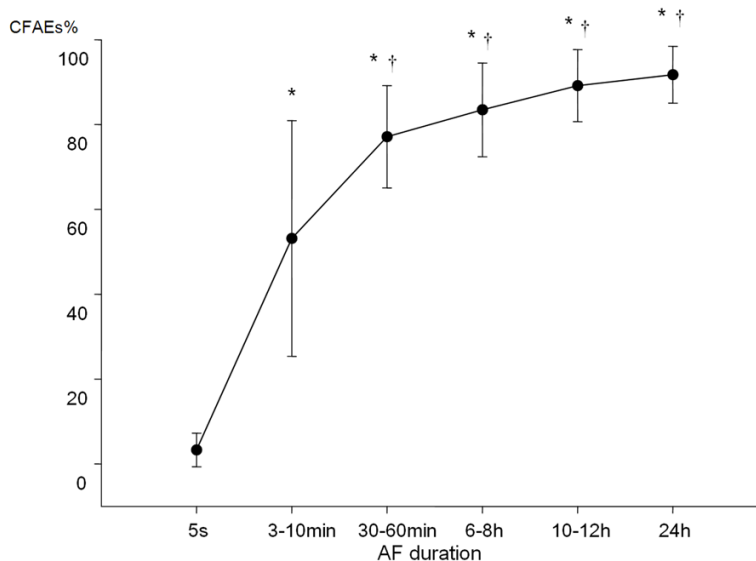


Figure 5. Changes in potentials in PVs with prolonged atrial fibrillation. LA = left atrium, SP = single potential, DP = double potential, FP = fragmented potential. * $P < 0.05$ compared with the occurrence of atrial fibrillation; † $P < 0.05$ compared with atrial fibrillation lasting for 3-10 min.

within 15 min. Propafenone was then infused at $4 \text{ mg} \cdot \text{kg}^{-1} \cdot \text{h}^{-1}$ until termination of AF. All cardiac electro signals were recorded for subsequent analysis.

After administration of propafenone, the average of 200 consecutive intervals in the LA and LSPV were recorded every 5 min as AFCL. The percentage of CFAEs was measured in the range of more than 300 consecutive activa-

tions during baseline, after AFCL was increased by 40 and 80 ms, and 16 s prior to cardioversion [16].

Statistical analysis

Statistical analysis was performed with SPSS version 10.0 (IBM Corp., NY, USA). Data are shown as mean \pm standard deviation. ANOVA was used for multiple-group comparisons, followed by a Bonferroni-corrected t-test. A two-tailed value of $P < 0.05$ was considered as significant.

Results

Establishment of a sustained AF model

Sustained AF was successfully induced in all of the eight goats. After rapid pacing for $20.2 \pm 8.1 \text{ h}$, and then 2.2 ± 1.0 , 5.3 ± 1.8 , and 7.3 ± 2.6 days, the duration of AF paroxysms increased from 3 to 10 min, 30 to 60 min, 6 to 8 h, and 10 to 12 h, respectively. After a mean of 9.3 ± 4.6 days' (range, 6-16 days) stimulation, AF lasted for more than 24 h spontaneously in all goats.

Development of CFAEs within PVs and the LA in the process of AF becoming stable

Figure 3 shows an example of changes in electrogram configuration with different stage at the LA. Pulmonary vein potentials (PVPs), which could not be detected in sinus rhythm, were unmasked by LA pacing. With a sustained increase in duration of induced AF, electrograms in the LA became fragmented and the ratio of CFAEs in the PV became higher (**Figures 4 and 5**). When induced AF lasted for more than 24 h, it almost became sustained ($3.3\% \pm 4.0\%$ vs. $91.8\% \pm 6.7\%$, onset of AF vs. AF lasted for 24 h, $P < 0.05$) in PVs and disappeared

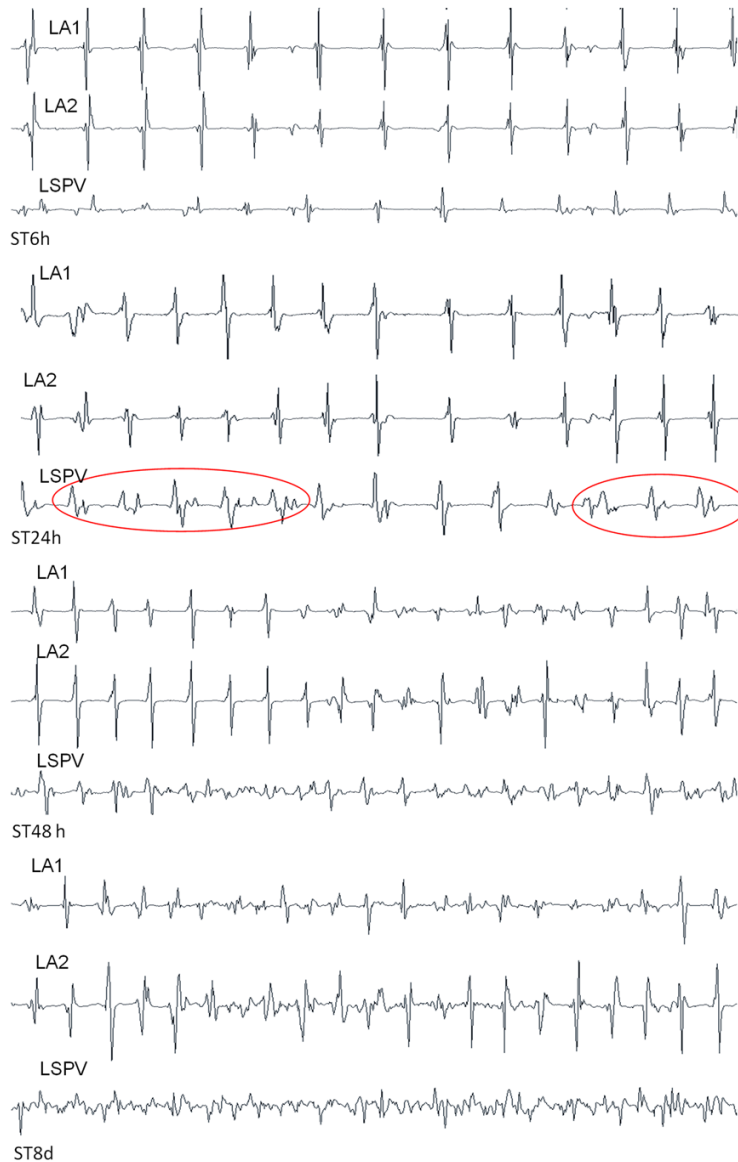


Figure 6. Example of CFAEs in the PVs in paroxysmal and sustained AF. Top panel: CFAEs in the PVs were found in the first episode of induced AF, and they lasted for only several seconds. Bottom panel: induced AF lasted for more than 24 h and CFAEs in PVs became almost sustained. LA = left atrium; LIPV = left inferior pulmonary vein; LSPV = left superior pulmonary vein.

before AF terminated automatically (**Figures 7 and 8**).

Atrial electrical remodeling in the process of AF becoming stable

During baseline, the mean AERP during regular pacing with intervals of 500, 400, 300, and 200 ms was 140.3 ± 15.2 , 141.6 ± 16.8 , 146.0 ± 18.8 , and 136.3 ± 13.0 ms, respectively. After 48 h of stimulation, the AERP was shortened to 60.3 ± 15.7 , 61.3 ± 23.1 , 61.0 ± 16.8 ,

and 66.7 ± 15 ms, respectively ($P < 0.05$ vs. baseline, **Table 1**). The AERP could no longer be measured after 48 h because, by that time, premature stimulation induced long-lasting paroxysms of AF that seriously hampered measurement. When induced AF lasted for 3-10 min, the mean RP_{AF} was 90.5 ± 13.2 ms and the mean AFCL was 98.3 ± 11.0 ms. On the first day that AF became persistent, the RP_{AF} and AFCL had further shortened to a mean of 63.0 ± 4.8 ms ($P < 0.05$ vs. AF lasted for 3-10 min) and 84.9 ± 5.2 ms ($P < 0.05$ vs. AF lasted for 3-10 min), respectively (**Table 2**). At the beginning of the stimulation, the AFCL in LA and AFCL in PV were 152 ± 29.0 and 146.7 ± 24.6 ms, respectively. When AF lasted for 24 h, the AFCL in LA and AFCL in PV were 84.9 ± 5.2 and 74.7 ± 6.2 ms ($P < 0.05$), respectively (**Figure 2**).

Comparison of CFAEs between the LA and PVs in development of AF

CFAEs in PVs were found in the first 24 h of LA stimulation, whereas there are almost a single potential in the LA (**Figure 6**). With prolongation of the stimulated duration, CFAEs increased faster in PVs than in the LA. The ratio of CFAEs in PVs was always higher than that in the LA at any stage of process of induced AF (**Figure 7**). When AF lasted for 24 h, the ratio of CFAEs in PVs was $91.8\% \pm 6.7\%$, the ratio of CFAEs in LA was $78.7\% \pm 4.6\%$ ($P < 0.05$). CFAEs in PVs lasted longer than in LA until AF terminated (**Figure 8**).

Termination of persistent AF

In all goats, persistent AF lasting more than 24 h could be cardioverted pharmacologically by infusion of propafenone (31.5 ± 15.8 mg). The

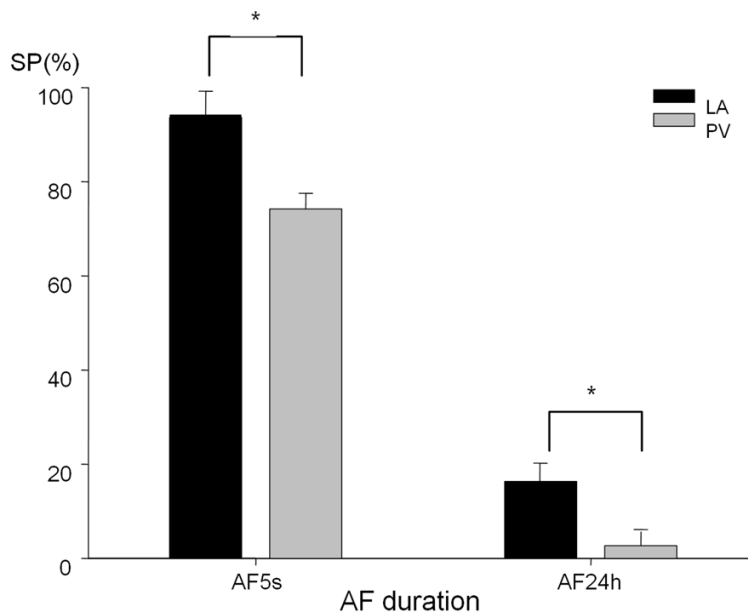


Figure 7. Ratio of single potential in the LA and PVs with prolonged atrial fibrillation. SP = single potential; LA = left atrium; PV = pulmonary vein.

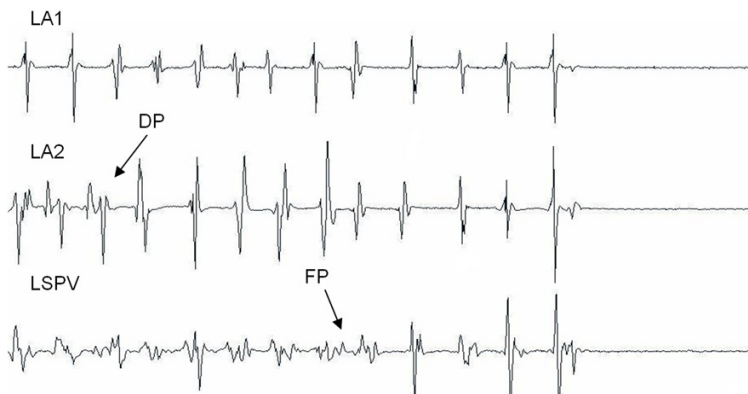


Figure 8. CFAEs in the PVs and LA disappeared before AF was terminated automatically. LA = left atrium, LA1 and LA2 indicate three different electrodes, which were sutured to different positions of the free wall of the left atrium. LSPV = left superior pulmonary vein.

mean time to cardioversion was 32 ± 16 mins. Before the termination of persistent AF, the CFAEs disappeared both in LA and PVs (**Figure 8**).

Changes in electrograms in the LSPV and LA by propafenone

After treatment with propafenone, the ratio of single potentials *increased gradually* in the LA and LSPV accompanied by prolonged AFCL ($P < 0.05$, **Table 3**). Double potentials and frag-

mented potentials in the LA converted to single potentials earlier than those in the LSPV. The mean time that double potentials and fragmented potentials disappeared with termination of AF was 5 s in the LA and 3 s in the LSPV ($P > 0.05$). Until AF was terminated, the LSPV had higher ratios of double potentials and fragmented potentials than did the LA (**Table 4**).

Discussion

Main findings

CFAEs in the PVs and LA increased with the process of atrial electrical remodeling and prolongation of the duration of AF. CFAEs were area-specific. Only after the CFAEs disappeared in PVs, could AF terminated.

CFAEs may be related to atrial electrical remodeling

The mechanism underlying CFAEs is not well understood. Multiple mechanisms have been hypothesized to explain CFAEs, including tissue anisotropy [1, 12, 17], colliding wave fronts [18], and conduction slowing at pivotal points of reentrant circuits [19]. Gerstenfeld et al. [12] found that the most common pattern of activation in regions of

CFAEs was that of wave front collision (47%) and short-radius reentry (38%).

CFAEs may result from electrical remodeling. Electrical remodeling creates conditions that are favorable to multiple-circuit reentry, which may then become a mechanism of CFAEs formation [20]. For example, the widening of the excitable gap ($AFCL - RP_{AF}$) will increase the probability that wave fronts encounter fully excitable tissue and decrease the likelihood of wave breaks and formation of new wavelets.

Table 1. Changes in AERP during the different durations of a stimulus

BCL	500 ms	400 ms	300 ms	200 ms
Baseline	140.3 ± 15.2	141.6 ± 16.8	146.0 ± 18.8	136.3 ± 13.0
St for 12 h	86.8 ± 12.7*	91.0 ± 11.5*	92.8 ± 12.6*	96.5 ± 6.7*
St for 24 h	70.8 ± 8.0*	72.3 ± 15.1*	71.3 ± 12.9*	75.8 ± 13.5*
St for 48 h	60.3 ± 15.7*	61.3 ± 23.1*	61.0 ± 16.8*	66.7 ± 15.0*. [†]

Data are shown as mean ± SD (n = 8). *P<0.05 vs. baseline; [†]P<0.05 vs. BCL = 500 ms. BCL = basic cycle length; St = stimulation.

Table 2. Changes in AFCL and RP_{AF} in the process of AF becoming stable

	AF duration				
	3 to 10 min	30 to 60 min	6 to 8 h	10 to 12 h	24 h
AFCL	98.3 ± 11.0	94.1 ± 10.6	86.9 ± 5.9*. [†]	85.7 ± 4.5*. [†]	84.9 ± 5.2*. [†]
RP _{AF}	90.5 ± 13.2	81.0 ± 14.1*	66.5 ± 3.4*. [†]	64.5 ± 3.0*. [†]	63.0 ± 4.8*. [†]

Data are shown as mean ± SD (n = 8). *P<0.05 vs. AF lasting for 3-10 minutes; [†]P<0.05 vs. AF lasting for 30-60 minutes.

Table 3. Changes in AFCL in the LA and LSPV after propafenone administration

	Baseline	AFCL + 40 ms	AFCL + 80 ms	Pre-cardioversion
LA	107 ± 7	147 ± 4*	185 ± 8*	225 ± 34*
LSPV	95 ± 9 [#]	135 ± 2*. [#]	167 ± 7*. [#]	211 ± 44*

Data are shown as mean ± SD (n = 8). *P<0.05 compared with baseline; [#]P<0.05 is a comparison between the LA and the LSPV indifferent period of AFCL prolongation.

Electrophysiological remodeling has been well described in experimental studies [13, 16]. Shan et al. [13] observed the AERP and AFCL during the transition from paroxysmal to persistent AF. They found fractionation of fibrillation electrograms increased with a decrease in atrial refractoriness and AFCL until AF became sustained. They also showed that administration of the class IC drug cibenzoline led to prolongation of AFCL and a reduction in the percentage of fragmentation [16]. Knecht et al. [21] reported that pharmacologically induced autonomic blockade resulted in an increase in AFCL exceeding 6 ms and a decrease in CFAEs. Rostock et al. [22] showed that a shortening of >10 ms in AFCL precedes the occurrence of CFAEs. In the present study, when the increase of stimulus intensity, the proportion of CFAE in both LA and PVs rise along with AERP shortened gradually. But when AF was sustained, AFCL in PVs decreased more rapidly than that in the LA (**Figure 2**). CFAEs increased more rapidly in PVs than in the LA. These findings indicated that the amount of CFAEs may be related

to the amplitude of variation of AERP and AFCL.

CFAEs are area-specific

CFAEs are area-specific. Nademanee et al. [1] divided the right and left atria into nine areas, they found that the CFAEs distributed differently and confirmed relatively in these regions. Some areas, such as PVs and pericardial adipose tissue with CFAEs, potentially represent AF substrate-sites. The architecture of the PV is different from other atrial sites. Multilayered muscles spread from the LA into the proximal PVs and fibrous tissue disseminates in myocardial clusters in the distal PVs [23]. These tissues have a different refractory period, and activation can be divided into many micropathways. A higher proportion of CFAEs at PVs may be the

result of dispersion of refractoriness between myocytes of PV's and the surrounding tissue, or it may be due to expression of local reentry within PVs. Tissue anisotropy may be the main mechanism of CFAEs induced by pericardial adipose tissue. Ashihara et al. [24] found that heterogeneous fibroblast proliferation in the myocardial sheet may be responsible for the genesis of CFAEs. Another study showed that the local CFAEs area has a significant relation between with regional pericardial fat volume [25]. Additionally, pericardial adipose tissue-based LA ablation can significantly decrease the CFAEs burden [26]. These findings further indicate CFAEs are area-specific and are closely related with pericardial adipose tissue.

The present study showed that the proportion of CFAEs in PVs was always higher than that in the LA at any stage of process of induced AF. There are at least four possible reasons for this finding. 1) A shorter AFCL in PVs after sustained AF may contribute to more CFAEs. 2) PVs have multilayered muscles spreading from the LA

Table 4. Effects of propafenone on the percentage of potentials in the LA and LSPV

Morphology	Baseline		AFCL + 40 ms		AFCL + 80 ms		Pre-cardioversion	
	LA	LSPV	LA	LSPV	LA	LSPV	LA	LSPV
SP	68 ± 6	46 ± 9 [#]	78 ± 5 [*]	63 ± 7 ^{*,#}	83 ± 4 [*]	74 ± 6 ^{*,#}	89 ± 6 [*]	80 ± 3 ^{*,#}
DP	16 ± 3	22 ± 5 [#]	11 ± 4 [*]	16 ± 3 ^{*,#}	9 ± 3 [*]	11 ± 3 ^{*,#}	5 ± 3 [*]	9 ± 1 ^{*,#}
FP	16 ± 3	31 ± 7 [#]	11 ± 3 [*]	21 ± 4 ^{*,#}	8 ± 2 [*]	15 ± 3 ^{*,#}	6 ± 1 [*]	11 ± 2 ^{*,#}

Data are shown as mean ± SD (n = 8). SP = single potential, DP = double potential, FP = fragmented potential. *P<0.05 compared with baseline; [#]P<0.05 is a comparison between the LA and the LSPV in the same period of AFCL prolongation.

into the proximal PVs, and distal PVs have disseminated myocardial clusters bordered by fibrous tissue [27, 28]. The complex organizational structure of PVs and the PV-LA junction may have contributed to our finding. 3) GP, which are located around the antrum of the PVs, may improve maintenance of CFAEs in PVs. 4) Pericardial adipose tissue secretes various inflammatory mediators and thus promotes the fibrotic cascade [29], resulting in an increase in CFAEs.

Effects of PVs CFAEs on the maintenance of AF

The PVs play an important role in clinical AF. CFAEs are often present during AF in PVs in humans. Radiofrequency catheter ablation of PVs eliminates these potentials, resulting in successful treatment of AF [1, 30, 31].

Whether development of CFAEs in PVs is responsible for the onset of stable AF is unclear. Chen et al. [32] studied changes in electrical activity of PV cardiomyocytes in dogs that were subjected to 6 to 8 weeks of atrial tachycardia. They observed a high prevalence of spontaneous early and delayed after depolarization in isolated PV cells. However, Cha et al. [33] did not observe after depolarization in the PV in coronary-perfused dog hearts that were subjected to 7 days of atrial tachycardia. Differences in after depolarization behavior are possibly related to the effect of cell isolation on PV action potentials. Cell isolation has potentially significant effects on ionic current properties [34].

Wu et al. [9] studied dogs with sustained AF, which was induced by chronic rapid atrial pacing. They observed a dominant frequency gradient during AF, where LA activity was faster than right atrium (RA) and CFAEs occurring in the PV and ligament of Marshall regions. Computerized mapping of the thoracic veins in this model

showed CFAEs in the PVs during AF [35]. Park et al. [11] recently showed that radiofrequency ablation encircling the PVs, ligament of Marshall, and superior vena cava terminates sustained AF induced by chronic rapid atrial pacing and prevents sustained induction of AF. Lee et al. [36] performed high-density epicardial mapping of the right superior pulmonary vein to obtain electrophysiological characterization of the PV-LA junction. They found that LA/PV pacing or programmed electrical stimulation increased functional conduction delay and circuitous activation patterns at the PV-LA junction, which created a substrate for reentry. These findings suggest that the thoracic veins might play a role in the maintenance of AF. Reentry and focal discharges may contribute to the development of CFAEs in the PVs [36]. However, Cha et al. [33] observed that, after 7 days of atrial tachycardia, resection of all PVs failed to alter inducibility of atrial tachyarrhythmia and did not significantly change the duration and cycle length of tachyarrhythmia. This finding indicates that a substrate for atrial tachyarrhythmia is present in the AT-remodeled atrium that does not necessarily require a contribution from the PVs.

Most previous animal model studies mapped PV activation only after persistent AF was induced. The mechanism of how CFAEs occur in the PVs has not been well studied. Zhou et al. [10] did not observe focal activation or reentrant wave fronts in the PVs during short-lived AF episodes in normal dogs. CFAEs could be found in the PVs during pacing-induced sustained AF. The present study showed that CFAEs in the PVs could be induced without chronic atrial pacing. This difference between studies may be due to a species difference. Dogs were used in Zhou et al.'s study [10], whereas goats were used in the present study. The present results favor the theory that the complex anatomic structure of PVs and the PV-LA connec-

tion contribute to the occurrence of CFAEs. With progressive shortening of the atrial refractory period and AFCL, more fibrillation waves coexist. AF becomes stable, and CFAEs in PVs are continuous.

Antiarrhythmic drugs (AAD) promote the fusion of fibrillatory wave fronts and decrease CFAEs [37]. The effect of AAD on AF may be owing to a widened excitable gap and prolongation of AFCL [11, 35, 38]. Because of widening of the excitable gap, fibrillation waves encounter tissue at a higher state of recovery of excitability. The wave fronts encounter fully excitable tissue more easily, and the wave breaks and new wavelets are hard to form. Shan et al. [13] also found that the excitable gap widened during the transition from paroxysmal to persistent AF. In the present study, after treatment with propafenone, which is a Na⁺ channel blocker commonly used for pharmacological cardioversion of AF, the proportion of CFAEs gradually decreased in the LA and LSPV along with the prolongation of AFCL. In the moment before the termination of atrial fibrillation, CFAEs in PVs disappear finally (**Figure 8**). It indicates that PVs may play an important role in the maintenance of AF.

Study limitations

There are several limitations in the study. First, only left PV and LA electrograms were analyzed. Other areas which CFAEs were frequently observed such as septum, the Bachmann bundle, crista terminalis and superior vena cava-right atrial junction [1] were not included. Secondly, only the AERP during AF in LA were analyzed. Because the PV electrograms were notoriously complex, could not be measured with the current method. However, the previous studies have suggested that the AERP in pulmonary vein would decrease along with the atrial electrical remodeling [31, 39].

Conclusion

CFAEs is area-specific and increased with atrial electrical remodeling. CFAEs in the PVs may play an important role in the maintenance of AF.

Disclosure of conflict of interest

None.

Abbreviations

AF, atrial fibrillation; CFAE, complex fractionated atrialelectrogram; PV, pulmonary vein; LA, left atrium; PVP, pulmonary vein potential; LSPV, left superior pulmonary vein; LIPV, left inferior pulmonary vein; AERP, atrial effective refractory period; RP_{AF}, atrial refractory period during AF; AFCL, atrial fibrillation cycle length; SP, single potential; DP, double potential; FP, fragmented potential; GP, ganglionated plexi.

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