

Atrial and ventricular electrical and contractile remodelling and reverse remodelling due to chronic pacing-induced atrial fibrillation in horses: preliminary results

Atriale en ventriculaire elektrische en contractiele “remodelling” en “reverse remodelling” van chronisch “pacing”-geïnduceerde atriale fibrillatie bij paarden: eerste resultaten

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Abstract

In humans, electrical and contractile reverse remodelling following restoration of sinus rhythm (SR) after a prolonged period of spontaneous atrial fibrillation (AF), requires several weeks. There is little known about this phenomenon in horses.

In the present study, six healthy horses were instrumented with a neurostimulator and a pacemaker to maintain AF for four months by intermittent burst pacing and to study atrial and ventricular electrophysiology. AF became persistent in all horses after two to six weeks of burst pacing. Before, during and after the AF period, parameters, such as the atrial fibrillation cycle length, the right atrial and ventricular refractory period and vulnerability, such as inducing atrial arrhythmias, atrial tachyarrhythmias or maintaining AF, were determined. Two-dimensional echocardiography was used to measure atrial and ventricular contractility expressed as fractional shortening and size expressed as diameter and area.

In two of the six horses, the procedure was discontinued due to an increased threshold (1 horse) and due to infection at the level of the pacemaker pocket (1 horse). In the four remaining horses, significant electrical and contractile remodelling compared to baseline values was observed from 48 hours onwards after AF induction. Upon restoration of SR with quindine sulfate, all electrical and contractile values returned to normal within one to two months. No ventricular remodelling was observed.

Four months of pacing-induced AF resulted in electrical and contractile remodelling and reverse remodelling. The results suggest that pacing-induced chronic AF does not cause permanent damage and suggest that a resting period of six to eight weeks before returning to training might be beneficial.

SAMENVATTING

Na een langdurige periode van atriumfibrillatie neemt de elektrische en contractiele herstelperiode bij de mens meerdere weken in beslag. Momenteel is er maar weinig bekend over dit fenomeen bij het paard.

In de voorliggende studie werden bij zes paarden een neurostimulator en een pacemaker ingepland om atriumfibrillatie (AF) te kunnen induceren en te onderhouden via een “burst-pacing”-mechanisme; en om de atriale en ventriculaire elektrofysiologie te kunnen bestuderen. Bij alle paarden ontstond persisterende AF na “burst pacing” gedurende twee tot vier weken. Parameters, zoals de atriale fibrillatiecyclus, de rechter atriale en ventriculaire refractaire periode en de gevoeligheid van het hart voor ritmestoornissen, zoals atriale aritmieën, atriale tachyaritmieën of AF, werden zowel voor en tijdens een AF-periode van vier maanden en gedurende een herstelperiode van twee maanden opgevolgd. De

atriale en ventriculaire contractiliteit uitgedrukt in fractionele “shortening” en de grootte van de hartcompartimenten werden opgevolgd via 2D-echocardiografie.

Bij twee van de zes paarden kon de procedure niet volledig opgevolgd worden ten gevolge van te hoge drempelwaarden (1 paard) of door een infectie ter hoogte van de “pocket” (1 paard). Bij de andere vier paarden konden er reeds na 48 uur van AF significante verschillen gemeten worden ten opzichte van de basale waarden. Tijdens de herstelperiode keerden alle waarden na één tot twee maanden naar de basale waarden terug.

In dit onderzoek wordt aangetoond dat geïnduceerde chronische AF, elektrische en contractiele veranderingen teweegbrengt ter hoogte van het hart bij het paard maar dat deze veranderingen omkeerbaar zijn eens het sinusritme hersteld is. Deze resultaten suggereren dat er geen permanente schade veroorzaakt wordt door vier maanden pacing-geïnduceerde AF en dat een rustperiode van zes tot acht weken gunstig zou zijn alvorens het werk te hervatten.

INTRODUCTION

Atrial fibrillation (AF) is clinically the most important supraventricular arrhythmia in horses (Deem and Fregin, 1982; Reef et al., 1988; Reef and McGuirk, 2002). AF can cause exercise intolerance, exercise-induced epistaxis, respiratory disease, sudden weakness, syncope or death due to ventricular dysrhythmias, which can only be detected with an electrocardiogram (Deem and Fregin, 1982; Reef and McGuirk, 2002, Verheyen et al., 2013). However, AF is often an incidental finding with no clinical complaints of performance loss that might already have existed for several weeks, months or potentially years.

In the human literature, AF has been described as a self-promoting electrical disease due to electrical, contractile and structural remodelling. As a result, successful pharmacological or electrical treatment becomes more difficult the longer the AF lasts (Alessie et al., 2002; Gold et al., 1986; Schotten et al., 2003). With a neurostimulator, it is possible to induce AF in horses. With a pacemaker and ultrasound, it is possible to follow up the electrophysiological and contractile changes that appear during an AF period and during the recovery period (after restoration of sinus rhythm). Initially, it is necessary to frequently stimulate the atrium with a neurostimulator in order to maintain AF; however, after an unknown period of time, AF becomes persistent. A similar follow-up of electrophysiological and contractile remodelling and reverse remodelling of short-term AF (i.e. seven days) has already been described in healthy horses (De Clercq et al., 2008). In that study, significant atrial electrical and contractile remodelling were observed from four and twelve hours onwards, respectively. After restoration of sinus rhythm (SR), reverse remodelling was completed within two days. These data suggest that early conversion of AF might be beneficial for the success rate, and that horses successfully treated for AF, only require a short resting period before returning to training. However, because many horses in clinical circumstances suffer from an AF duration of more than seven days, information about electrical and contractile remodelling and reverse remodelling after long-term AF is valuable.

In this study, electrophysiological and two-dimensional echocardiographic changes in the atrium and the ventricle during a four-month-period of induced AF were studied. In addition, the reversibility of the alterations was evaluated after permanent restoration of SR.

MATERIALS AND METHODS

The study was performed at the Ghent University (Belgium) and was supported by the Special Research Fund, Ghent University (Belgium). This research was approved by the ethics committee of the Faculty Veterinary Medicine, Ghent University (Belgium) (ECnr: 200235).

Animal and study protocol

Six healthy, untrained trotter horses with a mean age of 4.6 ± 0.5 years, a mean weight of 485 ± 24 kg and a mean height at the withers of 156 ± 5 cm, were included in the study. Clinical examination, electrocardiography and echocardiography, including two-dimensional, M-mode and Color flow Doppler, were performed by an equine internal medicine specialist with advanced skills in cardiology (DDC) to exclude underlying heart disease.

In all horses, a dual chamber pacemaker (Thera D(R), Medtronic, Minneapolis, MN) and a neurostimulator (Soletra 7426, Medtronic) were implanted, and an induced AF period was started as previously described to determine electrophysiological parameters before the AF period (thus in SR), during the AF period of four months and during the recovery period in SR (van Loon et al., 2001 and 2002; De Clercq et al., 2008). All horses were treated with quinidine sulfate for cardioversion to SR.

With the neurostimulator, intermittent burst pacing (three times threshold, 20 Hz, every two seconds) was performed until AF was induced. The dual chamber pacemaker allowed studying atrial and ventricular electrophysiology.

The study consisted of three periods: a baseline period (for the determination of the baseline values

(Figures 1, 2 and Tables 2, 3), a four-month AF period and a two-month recovery period. An overview of the timing of all measurements during the different periods is given in Table 1. The measured values can be found in Tables 2 and 3 and in Figures 1 and 2. Measurements during the baseline period were repeated on five different days to obtain a mean value. During the AF period, electrophysiological and echocardiographic measurements were performed in AF and within twenty minutes after spontaneous restoration of SR. When SR did not restore spontaneously, ventricular effective refractory period (VERP) and echocardiographic measurements were only performed in AF as atrial effective refractory period (AERP) can only be determined during SR. AF became persistent after two (horse 1 and 3), four (horse 2) and six weeks (horse 4). From the moment all echocardiographic and/or electrophysiological measurements were obtained in SR, the pulse generator was immediately switched on again for further maintenance of AF. After a certain period of time, AF was self-entertained and was thus persistent at the end of the four-month AF period, as in all horses. Subsequently, a pharmacological cardioversion with quinidine sulfate (QS) by a nasogastric tube (22mg/kg every 2 hours) was necessary to perform the next step of the study (i.e. the follow-up after restoration of sinus rhythm). The total cumulative dose of QS administered in a two-hour increments necessary to convert to SR ranged between 22 - 66 mg/kg.

Electrophysiological measurements

A programmed electrical stimulation test was performed in the atrium and the ventricle at three times baseline threshold amplitude for stimulation. During pacing with a fixed pacing interval (S1-S1), an extra stimulus (S2) was introduced with a coupling interval (S1-S2) below the expected refractory period. The coupling interval was then prolonged in steps of 8 ms until capture of the extra stimulus occurred, i.e. the atrial or ventricular S2 was followed by a P wave or QRS complex on the surface base-apex ECG, respectively. The longest S1-S2 interval without capture in the atrium or the ventricle was taken as AERP or VERP. As AERP changes with heart frequency, it was measured at pacing cycle lengths (PCL) of 1000 ms (60 beats/min), 800 ms (75 beats/min), 500 ms (120 beats/min) and 333 ms (180 beats/min). VERP was also measured at PCL of 1000, 800 and 500 ms. Two minutes of basic pacing was performed before starting to measure ERP (van Loon et al., 2002, De Clercq et al., 2008). At each PCL, AERP and VERP were determined three times to obtain a mean value. The atrial fibrillation cycle length (AFCL) was measured as the average time between two successive atrial depolarizations on a ten-second-intra-atrial electrogram.

The presence of arrhythmias during the AF induction period was evaluated via a standard base-apex electrocardiogram.

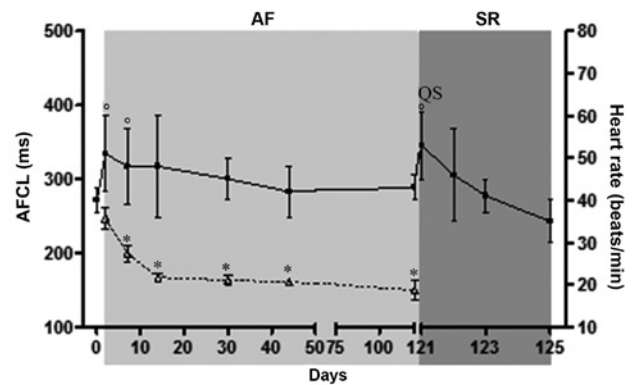


Figure 1. Mean (\pm SD) atrial fibrillation cycle length (AFCL; Δ -) and heart rate (HR; \blacksquare -) at baseline, during the AF period (light grey) and during recovery time (dark grey) in four horses. QS: treatment with quinidine sulfate. * = AFCL significantly different compared to the first AFCL measurement in the AF period ($p < 0.05$), \circ = significantly different compared to baseline measurement ($p < 0.05$).

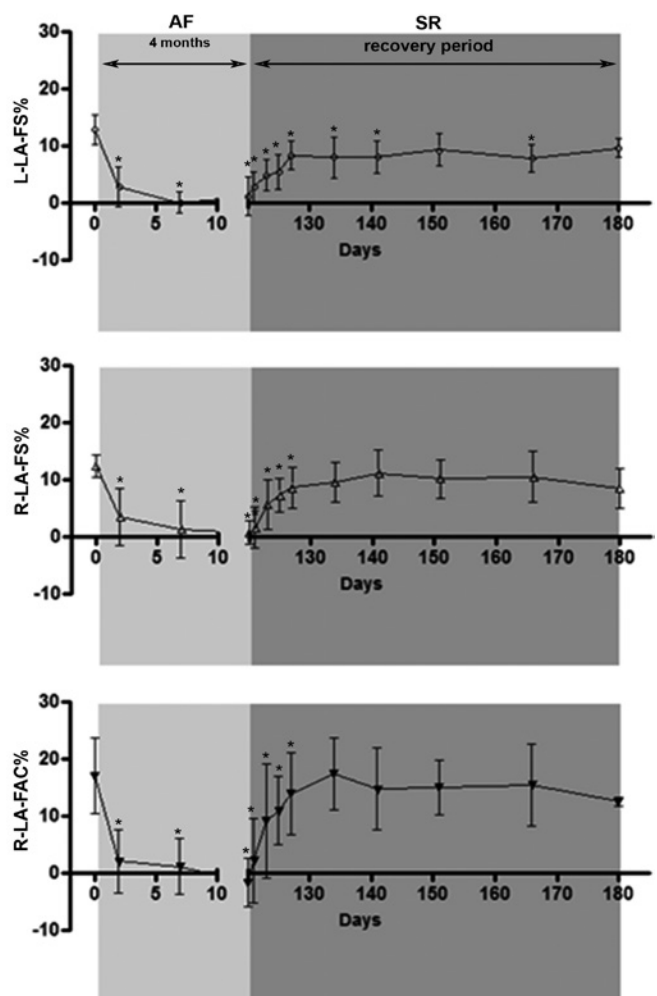


Figure 2. Left atrial fractional shortenings (FS%) (mean \pm SD) measured from the left (LLA-FS%) and from the right (LA-FS%) echocardiographic view and fractional area change (FAC%) at baseline during the AF period (light grey) and during recovery time (dark grey). * = significantly different compared to baseline.

Table 1. Overview of the timing of all measurements during the different periods. AF period is represented in light green and the recovery time period in dark green.

	Baseline	2d	1w	2w	4w	6w	4m	After QS	1d	3d	5d	1w	2w	3w	1m	1.5m	2m
Heart rhythm	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Heart rate	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
AFCL		+	+	+	+	+	+										
AERP	+	+				+	+	+	+	+	+	+	+	+	+		
Echocardiography	+	+	+				+	+	+	+	+	+	+	+	+	+	

QS: quinidine sulfate

d: days, w: weeks, m: months

Table 2. Mean (\pm SD) atrial effective refractory period (AERP) at pacing cycle length of 1000, 800, 500 and 333 ms and ventricular refractory period (VERP) at pacing cycle length of 1000, 800 and 500 ms during the AF period (light green) and during the recovery time period (dark green) in four horses.

	ERP 1000	AERP 800	AERP 500	AERP 333	VERP 1000	VERP 800	VERP 500
Baseline	256 \pm 22	254 \pm 24	249 \pm 28	226 \pm 23	358 \pm 11	322 \pm 15	251 \pm 18
2d	185 \pm 37*	184 \pm 38*	193 \pm 32*	182 \pm 27*	400 \pm 35*	349 \pm 41	258 \pm 45
1w	163 \pm 23*	168 \pm 21*	171 \pm 24*	165 \pm 28*	374 \pm 31	333 \pm 24	253 \pm 14
After QS	219 \pm 16*	225 \pm 30	248 \pm 30	148 \pm 08*	388 \pm 35	372 \pm 33*	276 \pm 36
1d	241 \pm 13	249 \pm 10	243 \pm 05	224 \pm 09	401 \pm 21*	346 \pm 14	251 \pm 19
3d	243 \pm 21	247 \pm 14	239 \pm 06	205 \pm 07	395 \pm 26*	355 \pm 13*	266 \pm 10
5d	250 \pm 21	251 \pm 17	245 \pm 07	208 \pm 10	395 \pm 21*	354 \pm 09	268 \pm 08
1w	248 \pm 13	246 \pm 14	242 \pm 07	206 \pm 07	383 \pm 30	345 \pm 23	262 \pm 21
2w	257 \pm 16	255 \pm 11	245 \pm 12	212 \pm 09	396 \pm 24*	365 \pm 20*	265 \pm 09
3w	255 \pm 14	255 \pm 07	241 \pm 11	214 \pm 13	395 \pm 20*	353 \pm 18	259 \pm 12
1m	255 \pm 21	255 \pm 18	245 \pm 16	209 \pm 15	404 \pm 12*	356 \pm 13*	262 \pm 13
1.5m	264 \pm 13	260 \pm 07	255 \pm 09	215 \pm 09	392 \pm 16*	353 \pm 20	265 \pm 18
2m	260 \pm 15	259 \pm 14	246 \pm 17	214 \pm 13	390 \pm 23*	351 \pm 13	261 \pm 08

QS: quinidine sulfate

* = significantly different from baseline ($p < 0.05$)

d: days, w: weeks, m: months

Echocardiographic views and measurements

Echocardiographic views were obtained using standardized imaging techniques with a 2.5 MHz sector transducer at a depth of 30 cm (GE Vingmed CFM 800 SV). A single-lead electrocardiogram was recorded simultaneously. Recordings were stored digitally and on video tape for retrospective analysis.

Every echocardiographic variable was determined from five cardiac cycles to obtain a mean value. Measurements were performed in SR at baseline, during the AF period (if SR restored spontaneously), and during the recovery period (in SR). Cycles during and immediately after a second-degree atrioventricular block or a spontaneous atrial premature beat were excluded from analysis. During the AF period, measurements were performed in AF. Measurements were performed at an RR interval between 1333 to 1090 ms (or a heart frequency between 45 to 55 beats/min). This is the heart rate that is most frequently seen in horses with spontaneous AF.

Left atrial dimensions were measured on a left parasternal view and a right parasternal long-axis

view. Left atrial inner diameter was measured on a left parasternal view close to the annulus of the mitral valve (L-LAD). On a right parasternal long-axis view, internal diameters were measured from the interatrial septum, close to the mitral annulus, to the atrial free wall (R-LAD). In the same view, a cross-sectional area (R-LAA) was measured (De Clercq et al., 2008). For every cardiac cycle, four-time points were determined to perform all these measurements: point "p" was at the onset of the P wave; point "a" was during maximal atrial contraction indicated by smallest atrial volume, point "d" was at the end of the ventricular diastole at the moment the mitral valve had just been closed, and point "s" was at the end of ventricular systole. Due to the absence of a P wave and an atrial contraction, points "p" and "a" could not be determined during AF.

Left atrial fractional shortening (LA-FS%) and left atrial fractional change (LA-FAC%) were calculated as follows (Piotrowski et al., 2000):

$$LA - FS\% = \frac{LADp - LADa}{LADp} \times 100$$

$$LA - FAC\% = \frac{LAAp - LAAa}{LAAp} \times 100$$

On a standard M-mode of the ventricles, the left ventricular internal diameter, the interventricular septal thickness and the left ventricular free wall thickness were measured in diastole and systole; and the left ventricular fractional shortening was calculated. Color flow Doppler examinations of the mitral, tricuspid, aortic and pulmonic valves were performed. All ECG recordings were performed with the electrodes in a standard base apex position for horses. No cardiac enzyme activity was followed during this study.

Statistical analysis

Data are shown as mean ± standard deviation. First, a linear mixed model (univariate analysis of variance) was applied with ‘horse’ as random effect and ‘time’ as categorical fixed effect, at a significance level of 5%. If the effect of time was significant, a post hoc Dunnett’s test was performed to compare all time points to the baseline value at time zero to adjust for multiple comparisons, at a significance level of 5%. Heart rate and echocardiographic measurements of LA dimensions in SR and in AF were compared using a linear mixed model with ‘time’ and ‘rhythm’ (SR or AF) as fixed factors and ‘horse’ as the unit of repeated measure.

RESULTS

After pacemaker implantation, two horses were excluded from the study protocol due to an increased

threshold of the pacemaker lead; this lead was used for the electrophysiological measurements (1 horse), and one due to infection at the level of the pacemaker pocket (1 horse). In the first case, the pacemaker and the leads were removed; in the second case, the pacemaker leads and the pacemakers were removed and an antimicrobial treatment was started. Both horses recovered successfully. In the other four horses, AF could be successfully induced and maintained using the intermittent burst pacing protocol. AF became persistent after two (horses 1 and 3), four (horse 2) and six weeks (horse 4) of maintenance by burst pacing; thus, the horses became vulnerable for AF over time. All horses were successfully converted to SR after a total of two or three doses of QS at a two-hours’ interval. One horse (horse 2) showed widening of the QRS complex and tachycardia after the second dose of QS. In two horses (horse 1 and 4), an electrocardiogram showed that atrial premature complexes were present until three weeks after restoration of SR.

Electrophysiological measurements

Except for horse 2, atrial programmed electrical stimulation could be performed according to the predefined protocol. In horse 2, atrial threshold had risen above the maximum output levels of the pacemaker when SR was restored after four months of AF. There-fore, in horse 2, electrical measurements during the recovery period were performed with a temporary pacing catheter (Bipolar Intracardiac Electrode, USCI) using the same pacing protocol. In all

Table 3. Mean (±SD) echocardiographic measurements before, during the AF period (light green), and after restoration to sinus rhythm (dark green). L-LAD: left atrial diameter measured from a left parasternal long-axis view. R-LAD: left atrial diameter measured from a right parasternal long-axis view. R-LAA: left atrial area measured from a right parasternal long-axis view. d: end ventricular diastole, s: end ventricular systole.

	Left parasternal long-axis view	Left parasternal long-axis view	Right parasternal long-axis view	Right parasternal long-axis view	Right parasternal long-axis view	Right parasternal long-axis view
	LLADd	LLADs	LADd	LADs	LAA d	LAA s
Baseline	9.1±0.3	10.4±0.4	8.8±0.5	9.7±0.6	41.4±6.7	55.1±5.8
2d	8.2±0.9	9.9±0.8	8.4±0.8	8.7±0.8*	41.1±9.8	45.3±7.6*
1w	9.0±0.9	10.0±0.8	8.3±0.9	8.7±0.9*	41.2±8.5	45.7±6.7*
Just after QS	9.8±1.0*	10.6±0.8	9.1±0.9	9.8±0.8	50.0±11.6*	58.93±10.6
1d	9.5±0.8	10.5±0.7	9.3±1.0	10.0±0.9	49.3±10.5	59.7±10.6
3d	9.5±0.8	10.5±0.7	9.3±0.9	9.9±0.9	49.3±10.5	59.7±10.6
5d	9.4±0.7	10.4±0.7	8.9±0.7	9.5±0.8	47.3±7.2	57.2±8.6
1w	8.9±0.6	10.2±0.7	8.6±0.5	9.6±0.7	46.1±8.0	55.1±6.9
2w	9.1±1.0	10.4±1.1	8.6±0.7	9.6±0.9	43.86±6.9	55.1±8.8
3w	9.1±1.2	10.3±0.7	8.4±0.8	9.2±1.0	43.4±8.8	52.5±8.8
1m	8.9±0.9	10.2±0.7	8.5±0.7	9.4±0.9	42.7±6.5	53.4±5.3
1.5m	9.0±0.8	10.1±0.9	8.4±0.8	9.4±0.7	42.5±7.6	53.7±9.5
2m	9.0±0.9	10.1±0.8	8.5±0.8	9.24±0.9	41.6±9.2	53.6±9.2

QS: quinidine sulfate

* significantly different from baseline (p<0.05).

d: days, w: weeks, m: months

horses the threshold of the ventricular lead increased slightly during the AF period, which resulted in borderline stimulations (just below three times baseline threshold) during the ventricular programmed electrical stimulation studies.

Atrial fibrillation

After one day of maintained AF, the mean AFCL of the four horses was 243 ± 13 ms and decreased to 151 ± 15 ms after four months of AF. From two days onwards, there was a significant decrease of AFCL (Figure 1). A significantly increased heart rate was only observed during the first week of AF and on the day of quinidine treatment (Figure 1). There was no significant difference between the heart rate measured at the same time point in SR or in AF during the AF period (data not visible).

Atrial electrophysiology

At baseline, AERP was 256 ± 22 ms, 254 ± 24 ms, 249 ± 28 ms and 226 ± 23 ms at a pacing CL of 1000, 800, 500 and 333 ms, respectively. After one week of maintained AF, AERP had shortened significantly (Table 2). Quinidine treatment resulted in an immediate significant increase of AERP. One day was necessary to achieve AERP baseline values after the pharmacological treatment (Table 2). Shortening of AERP (during the AF-period) was associated with atrial premature beats, atrial tachycardia or a period of short-term AF. The presence of atrial premature beats, atrial tachyarrhythmias or short-term AF varied per individual horse.

Ventricular electrophysiology

At baseline, VERP was 358 ± 11 ms, 322 ± 15 ms and 251 ± 18 ms for a CL of 1000, 800 and 500 ms, respectively (Table 2). A significant increase of VERP for a CL of 1000 and 800 occurred during the recovery period. VERP measurements were never associated with ventricle tachycardia or ventricle fibrillation.

Echocardiographic measurements

Four months of AF did not result in a significant increase of R-LADd, L-LADd, R-LADs, L-LADs, R-LAAd and R-LAAs.

R-LA-FS%, L-LA-FS% and R-LA-FAC% had already decreased significantly within two days of maintained AF ($p < 0.0001$). After one week of AF, the atrial contractile function was almost completely absent. Values recovered within one to two months of restoration of SR (Figure 2).

The left atrial sizes measured during AF were comparable with the sizes measured in SR at the comparable time point (only measurements performed during AF are given in Table 3).

No significant changes were observed in inter-ventricular septal thickness, left ventricular free wall thickness, left ventricular internal diameters nor in left ventricular fractional shortening throughout the study. Color flow Doppler of all valves revealed no differences compared to baseline (there were no pathological valvular regurgitations present before, during or after the study).

DISCUSSION

In the present study, electrical and contractile remodelling was evaluated during an artificially induced chronic AF model in horses. The major findings of this study were: (1) pacing-induced AF resulted in atrial electrical and contractile remodelling; (2) AFCL decreased significantly during the first fourteen days of AF and remained significantly lower throughout the AF period; (3) two-dimensional ultrasound did not allow to demonstrate significant differences in atrial size; (4) electrical reverse remodelling after pharmacological conversion of sinus rhythm appeared faster than contractile reverse remodelling.

Horses with naturally occurring AF are frequently presented with subacute (two weeks) or chronic AF. In addition, these horses are commonly classified as 'lone' fibrillators, because echocardiography reveals no or only clinically non-relevant abnormalities. The purpose of the present study was to examine 'lone' AF in horses. Therefore, young and healthy horses were selected without abnormalities on clinical examination, electrocardiography at rest and during exercise and echocardiography including two-dimensional ultrasound, M-mode and Color flow Doppler. Clinical examination, electrocardiography during rest and exercise and echocardiography was performed by an equine internal medicine specialist with advanced skills in cardiology (DDC).

The burst pacing protocol was effective to induce and maintain AF in four out of six horses (two horses were left out of the study due to increased threshold and infection of the pacemaker pocket) even though two of the four remaining horses needed a period of pacing (horse 2: four weeks; horse 4: six weeks) before AF became persistent. Besides the measurements made during AF, brief periods of spontaneously restored SR allowed to make electrophysiological measurements during normal SR during the first eight days of the protocol in all horses without any administration of drugs. However, persistent AF appeared after 8, 28, 10 and 42 days for horse 1, 2, 3 and 4, respectively. The progressive increase in susceptibility for AF has also been shown in experimentally induced atrial fibrillation in ponies, goats and dogs (Morillo et al., 1995; Wijffels et al., 1995; van Loon et al., 2001). In the study by Morillo et al. (1995), a six-weeks' episode of rapid atrial pacing induced AF lasting for more than 15 minutes in 82% of the studied dogs; while

in a study by van Loon et al. (2002), six months of burst pacing resulted in persistent AF in only one of four research ponies. The fact that horses have more tendency to fibrillate than small animals, i.e. dogs, ponies, can be explained, at least in part, because horses have a larger atrial size. This explains why naturally occurring AF is more frequently encountered in horses than in ponies (Else and Holmes, 1971; Bertone and Wingfield, 1987; Detweiler, 1989). The old multiple wavelet re-entry theory of Moe (1962) partially explains why AF has more tendency to persist in larger atria. In this theory, Moe (1962) described that the presence of AF is dependent on the possibility of the atrium to maintain a critical number of re-entry circuits. The development therefore depends on the atrial diameter and the size of the re-entry circuits. This re-entry circuit size is expressed as the wavelength (WL) of re-entrant circuits and is defined by the AERP and conduction velocity (CV) ($WL = AERP \times CV$). AF develops more easily if the atrial diameter is larger and/or the wavelength is shorter; thus, if the AERP or the CV decreases.

In humans and dogs, AF is characterized by a rapid and irregular ventricular response. However, during atrial flutter, which is accompanied by a slower atrial rate, a higher ventricular response is often seen since more atrial impulses are transmitted to the ventricles probably due to less concealed conduction at the atrio-ventricular node (Hnatkova et al., 1998; Miller et al., 1999; King et al., 2002; Silbauer and Sulke, 2007). In the horses of the present study, only a significant increased heart rate during the first week of AF was observed.

The decrease in AFCL might be explained by an increase in the number of wavelets due to the shortening of the AERP with the increasing duration of AF (Moe, 1962).

In the horses of the present study, a significant increase in VERP during the AF period and during the recovery period was observed. During the study, there was a small increased threshold of the ventricular lead, whereby maximal pacemaker output was needed to reach three times threshold amplitude. This technical limitation might, in part, explain the increased VERP since borderline or submaximal power can affect VERP values.

A shortening of AERP and a reduction of atrial contractility during the first hours and days of AF induction has been observed in dogs, goats, horses and humans (Olsson et al., 1971; Cotoi et al., 1972; Wijffels et al., 1995; Franz et al., 1997; Schotten and Allessie, 2001; Allessie et al., 2002; Schotten et al., 2003; Schwarzwald et al., 2007; Thanigaimani et al., 2017). In an AF-induced model in goats, a new steady state of atrial refractory period appeared after two to three days and atrial contractile function was almost completely lost after two days (Wijffels et al., 1995). This shortening of AERP leads to an increased vulnerability for AF, which can eventually lead to persistent

AF (Wijffels et al., 1995). In a study by Allessie et al., (2002), reverse electrical remodelling was still completely reversible in goats after months or even years of AF. It took only a few days for AERP to become normal again after seven weeks of AF (Allessie et al., 2002). After a six-week AF period, atrial contractile function was recovered one month after conversion in human patients (Manning et al., 1994). Similar changes were observed during the present study in horses.

The occurrence of atrial premature beats after conversion, as observed in the horses of the present study, has also been described in humans (Maounis et al., 2001) and is also observed in some horses who were treated for naturally occurring AF. The pathophysiology of electrical and contractile remodelling is quite complex and is only partly understood, but calcium levels and the sensitivity of calcium channels to calcium play a major role. Atrial tachycardia induces intracellular calcium overload followed by a downregulation of the L-type calcium channels and a subsequent reduction of sarcolemmal calcium influx. This eventually results in a shortening of the AERP or a loss of the plateau phase of the action potential (Yue et al., 1997; Yue et al., 1999).

The short refractory period results in a reduction of wavelength, which increases the vulnerability for AF. However, this could not explain the subsequent decreased efficacy of cardioversion and maintenance of SR in chronically instrumented goats with prolonged AF duration, because atrial remodelling appears rather quickly (Wijffels et al., 1995). Possibly, one or more other factors may be involved in the increase in vulnerability for AF. One of these factors is loss of the atrial contractile function (Tieleman and Crijns, 2000; Thanigaimani et al., 2017; Thomas and Abhayaratna, 2017). In humans, this process also occurs due to alterations in cellular calcium handling (Sun et al., 1998). In addition, this contractile dysfunction may lead to atrial paralysis and an increase in compliance of the atria. Subsequently, this may cause dilatation of the atria during the course of several days to weeks of AF (Schotten and Allessie, 2001; Thomas and Abhayaratna, 2017). In the four horses of the present study, a rapid decline of AERP and a loss of atrial contractile function with an increase in vulnerability were also observed. In the artificial AF model used in the present study, the increase in atrial size was not significant in the four horses. AF became persistent in each horse necessitating cardioversion.

In humans, atrial contractile dysfunction has been suggested to delay the improvement in exercise intolerance after cardioversion, and may influence the recurrence of AF (Harjai et al., 1997; Khan, 2003; Schotten et al., 2003). It is not known whether this delay in exercise intolerance also applies for horses. However, in a study in horses with naturally occurring chronic AF (1.5-24 months), the atrial contractile function was restored seven weeks after conversion, although the atrial dimensions were still increased

compared to the reference values of normal horses (Decloedt et al., 2013).

LIMITATIONS OF THE STUDY

In this preliminary study, two of the six horses could not complete the study due to technical problems. Therefore, the results regard a very small study population. In addition, the study was not blinded and a control group was absent.

CONCLUSION

The results of this study indicate that, in the four healthy horses, experimentally induced AF brings about rapid electrophysiological and contractile remodelling. In each horse, self-entertaining and thus persistent AF appeared after one to six weeks of intermittent burst pacing. After pharmacological restoration to SR, rapid electrical reverse remodelling and a slower normalization of atrial contractility were observed within a few weeks. This study indicates that induced chronic, pacing-induced AF does not suggest permanent electrical alterations and that reverse remodelling is still possible after a chronic, pacing-induced AF period of four months.

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