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# Holter electrocardiography in dogs showing doxorubicin-induced dilated cardiomyopathy

[*Eletrocardiografia Holter em cães com cardiomiopatia dilatada experimental induzida pela doxorrubicina*]

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## ABSTRACT

Early identification of arrhythmias in dogs showing doxorubicin-induced cardiomyopathy was studied. Ten healthy dogs were assigned to groups A (n=5) and B (n=5). Dogs from group B were given doxorubicin 30mg/m$^2$ intravenously, every 21 days, until a cumulative dose of 180mg/m$^2$ or 240mg/m$^2$ was reached. Dogs from group A (used as control) were administered saline intravenously at the same group B intervals. As soon as myocardium dysfunction was observed in dogs from group B, determined by a shortening fraction below 20%, increased E-point to septal separation above 0.7cm, and increased end-systolic left ventricular volume index (61.4ml/m$^2$), a 24-hour Holter was recorded in all dogs from both groups. There was an increase of minimum heart rate (44.6%) and mean heart rate (41.7%) in animals from group B in comparison with the control animals. Either supraventricular or ventricular arrhythmias were observed, despite group B dogs showed higher occurrence of supraventricular arrhytmias. Holter monitoring is efficient in early determination of heart rate and cardiac rhythm alterations in dogs showing doxorubicin-induced myocardial dysfunction.

Keywords: dog, doxorubicin, eletrocardiogram, cardiopathy

## RESUMO

*O estudo consistiu na identificação precoce da ocorrência de arritmias em cães com cardiomiopatia dilatada experimental induzida pela doxorrubicina (DOX). Utilizaram-se 10 cães adultos, sadios, distribuídos nos grupos A (n=5) e B (n=5). O grupo B recebeu 30mg/m$^2$ de DOX, via intravenosa, a cada 21 dias, até a dose cumulativa de 180 ou 240mg/m$^2$. No grupo A (controle), administrou-se solução salina 0,9%, via intravenosa, nos mesmos intervalos do grupo B. Ao se evidenciar o quadro de disfunção miocárdica nos cães do grupo B, caracterizado pela fração de encurtamento menor que 20%, aumento da separação septal do ponto E acima de 0,7cm e aumento do índice volumétrico do ventrículo esquerdo ao final da sístole (61,4ml/m$^2$), realizaram-se os eletrocardiogramas por 24 horas. Os resultados demonstraram aumentos de 44,6% e 41,7% nas freqüências cardíacas mínima e média, respectivamente, e presença, com maior freqüência, de arritmias supraventriculares do que ventriculares nos animais do grupo B. Concluiu-se que o Holter é eficaz e demonstra, com precocidade e melhor definição, as alterações da freqüência e do ritmo cardíaco de cães com disfunção miocárdica induzida pela doxorrubicina.*

*Palavras-chave: cão, doxorrubicina, eletrocardiograma, cardiopatia*

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## INTRODUCTION

Doxorubicin chloride is a widely used antineoplastic agent from the anthracycline group, which is obtained by fermentation made by *Streptomyces peucetius* var. *caesius* fungi. It shows an oncolitic action in the treatment of leukemias, lymphomas, osteogenic sarcomas, breast cancer, ovarian carcinoma, gastric tumors and carcinoma of the lungs. This drug has been widely used in medicine and, nowadays, it has been used in some chemotherapy protocols in veterinary medicine (Susaneck, 1983; Postorino et al., 1989; Jacobs, 1996; Dias et al., 1997; Dagli, 2002).

Anthracycline drugs produce highly reactive free radicals that are able to disrupt DNA and cellular membranes, causing alteration of their functions. Such free radicals can induce lesions in the myocardium, characterizing a cardiotoxic effect, therefore, limiting the dose to be administered (Gwathmey and Davidoff, 1994; Dagli, 2002).

The recommended dose of doxorubicin for dogs is 30mg/m$^2$, given each 21 days, to maximize its antineoplastic actions and reduce the toxicity (Susaneck, 1983; Mauldin et al., 1992; Dias et al., 1997). It should be cautiously administered, since a severe vesicant action and local tissue necrosis can occur (Chabner et al., 1996).

Due to the severe cardiotoxicity of doxorubicin, it should be carefully used in veterinary medical practice. Cardiotoxicity in dogs (chronic toxicity) is related to cumulative doses over 250mg/m$^2$, which can lead to the development of cardiomyopathy and congestive heart failure. However, it has been experimentally observed that dogs develop dilated cardiomyopathy (DCM) and congestive heart failure when receiving much lower cumulative doses of doxorubicin up to 200mg/m$^2$. In some dogs, doses up to 90mg/m$^2$ may cause cardiac abnormalities (Mauldin et al., 1992; Jacobs, 1996).

The cardiotoxic effect is a syndrome characterized by congestive heart failure (CHF), electrocardiographic abnormalities, arrhythmias and even sudden death. Moreover, it could reveal during the treatment and for a long time after finishing the treatment (Mauldin et al., 1992; Jacobs, 1996).

The most common electrocardiographic findings associated with doxorubicin therapy include changes in ST segment and T wave, as well as EKG patterns, suggestive of enlargement of left atria and right and left ventricles, supraventricular arrhythmias, such as atrial premature complexes and atrial fibrillation. It is also possible to occur ventricular arrhythmias, intraventricular conduction disturbances and sinus tachycardia (Kehoe et al., 1978; Susaneck, 1983; Cowan and Jacobs, 1990; Crystal and Rush, 1991; Mauldin et al., 1992; Jacobs, 1996; Andrade, 2002; Silva and Camacho, 2005).

Electrocardiographic alterations in doxorubicin-induced cardiomyopathy can be early detected by ambulatory electrocardiography (Holter system), which is based on electrocardiographic recording for a long period, generally up to 24 hours. It is the most sensible non-invasive test that allows demonstrating transitory arrhythmias not detected by standard electrocardiography (Hall et al., 1991; Miller and Calvert, 1992; Goodwin, 1998; Mucha, 2001).

The aim of this research was to perform early diagnosis and characterization of arrhythmias in dogs with doxorubicin-induced cardiomyopathy.

## MATERIAL AND METHODS

Ten adult mongrel healthy dogs, males and females, were selected. Prior to the beginning of the experiment, all animals wee submitted to a detailed physical examination, besides a complete analysis of blood elements and biochemical profile (BUN, creatinine, alkaline phosphatase, aspartate aminotransferase, total protein, albumin, total bilirubin, and direct bilirubin). Standard electrocardiography and echocardiogram were also performed to rule out any kind of anatomic or functional abnormality. Dogs were randomly divided into two groups of five animals each. Group A (control) was composed of healthy dogs and dogs with doxorubicin-induced DCM were sampled only in group B.

Animals from group B were given doxorubicin 30mg/m$^2$, each 21 days, until a cumulative dose of 180 or 240mg/m$^2$ was reached. Such dose is known to cause myocardial dysfunction. The drug was slowly infused IV. Dogs from group A

were given 0.9% isotonic saline by the same route and at similar intervals of group B.

Prior to each administration of doxorubicin, an echocardiogram was performed only to determine the development of myocardial dysfunction, which was characterized by a decreased fractional shortening to values below 20%, increased E-point to septal separation above 0.7 cm, and increased end-systolic left ventricular volume index (61.4 ml/$m^2$).

Once myocardial dysfunction was detected, doxorubicin administration was discontinued and dogs from group B were submitted to ambulatory electrocardiography for 24 hours. It was used a Holter device[1], with two precordial leads (rV2 and V4), whose recordings were recorded in a standard cassette tape.

Three dogs from group B showed myocardial dysfunction when a cumulative dose of 180 mg/$m^2$ of doxorubicin was reached, while in the remaining animals, such dysfunction was observed when 240mg/$m^2$ of that drug was reached. The 24-hour EKG (Holter) was also performed in dogs from group A, after echocardiographic examination, in accordance with group B.

The Holter device was stored in a comfortable leather jacket, which allowed the dogs to walk freely, with no restriction to movements. The jacket was used to protect the cables that were attached to adhesive electrodes positioned on different parts of the chest and linked to the recorder. The device was turned on and set up to turn off 24 hours later. When recording was finished, the cassette tapes were decoded through a computerized system at the Holter Interpretation Service of the Instituto de Moléstias Cardiovasculares de São José do Rio Preto (São José do Rio Preto, São Paulo, Brazil).

The data were statistically analyzed by a simple Student t test with independent samples, two treatments and 5 replicates, at a level of probability of 5%.

## RESULTS

The Holter variables included heart rate (the lowest, mean and the highest), total number of QRS complexes, total number of supraventricular arrhythmias (sole, in couplets and as tachycardia), total number of ventricular arrhythmias (sole, bigeminism, in couplets and as tachycardia), asystole, depression or elevation of ST segment (Tables 1 and 2).

Table 1. Mean and standard deviation of minimum and maximum heart rates (bpm), number of QRS complexes, ectopic supraventricular and ventricular beats, number of asystole, and the longest pause (in seconds), according to ambulatorial 24-hour electrocardiography in dogs showing doxorubicin-induced cardiomyopathy (n=5) and of control dogs (n=5)

| | Control | Doxorubicin |
|---|---|---|
| min HR | 43.0±3.74 | 62.2±16.0* |
| Aver HR | 86.4±3.91 | 122.4±21.12* |
| max HR | 246.0±6.52 | 238.0±16.97 |
| # of QRS | 120586.4±5582.8 | 172631.80±30320.18* |
| SVE | _ | 2966.20±4441.0* |
| VE | _ | 200.4±237.96* |
| Asystole | 8.0±13.04 | 10±18.56 |
| Longest pause | 1.08±13.04 | 1.62±1.52 |

min HR=minimum heart rate; aver HR=average heart rate; max HR=maximum heart rate; # of QRS=number of QRS complexes; SVE=ectopic supraventricular beats; VE=ectopic ventricular beats.
*Statistically significant results in relation to data of same rows.

The lowest and mean heart rate showed significant alterations ($P<0.05$) between the groups, with the greatest means recorded in animals from group B. However, the mean values of the highest heart rate in such group did not show significant differences ($P>0.05$) when compared with the results of the control group (group A).

In relation to the number of QRS complexes, the mean values increased significantly ($P<0.05$) in animals submitted to the chronic treatment with doxorubicin, in comparison with animals from the control group.

[1]Holter Recorder, Model 90205, Space Labs, Washington/USA

Table 2. Absolute and mean values of ectopic supraventricular and ventricular beats, according to ambulatorial 24-hour electrocardiography in dogs showing doxorubicin-induced cardiomyopathy (n=5)

| | SVE | | | VE | | | |
|---|---|---|---|---|---|---|---|
| Animal | Isolated | Couplets | Tachycardia | Isolated | Bigeminism | Couplets | Tachycardia |
| 1 | 512 | 107 | 1582 | 294 | 3 | 33 | 20 |
| 2 | 243 | 3 | 1 | 3 | 0 | 2 | 0 |
| 3 | 669 | 178 | 623 | 343 | 0 | 46 | 5 |
| 4 | 8 | 0 | 0 | 41 | 0 | 5 | 0 |
| 5 | 77 | 1 | 6 | 17 | 0 | 1 | 1 |
| Mean±SD | 301.8±282 | 57.8±81 | 442.4±691 | 139.6±164 | 0.6±1 | 17.4±20 | 5.2±8 |

SVE=ectopic supraventricular beats; VE=ectopic ventricular beats; SD=standard deviation.

The animals from control group neither developed supraventricular nor ventricular arrhythmias, as determined by the 24-hour ambulatory electrocardiography. However, animals from group B showed frequent arrhythmias ($P<0.05$), mostly supraventricular rather than ventricular.

It was not determined statistical differences ($P>0.05$) between the related groups to the occurrence of asystole or pauses, longer than two seconds. In relation to ST segment, the 24-hour ambulatory electrocardiography (Holter system) did not reveal elevations or depressions in both groups.

## DISCUSSION

The chronic administration of doxorubicin chloride resulted in cardiotoxicity being the most significant side effect. Such effect can lead to cardiac dysfunction and the development of DCM, which is manifested by supraventricular and ventricular arrhythmias, myocardial failure, or both, similar to what was observed in dogs from group B, which was previously described by Kehoe (1978), Mauldin et al. (1992), Jacobs (1996), Kittleson (1998), Andrade (2002), Souza (2004) and Silva and Camacho, 2005.

Electrocardiographic monitoring for 24 hours allowed a continuous recording of the cardiac electrical activity, which made possible a better verification of the occurrence of arrhythmias. Sometimes they were transient in dogs suffering doxorubicin-induced cardiomyopathy. Therefore, they could not be promptly identified through standard electrocardiography (Mauldin et al., 1992; Kittleson, 1998).

Less than 1% of human subjects develop signs of cardiotoxicity when doses of up to 500mg/m$^2$ of doxorubicin are given (Lefrak et al., 1973). In this study, it was observed that signs of cardiac dysfunction were firstly developed by three dogs, when a cumulative dose of 180mg/m$^2$ was reached, followed by two animals when a cumulative dose of 240mg/m$^2$ was reached. This suggests that dogs are more sensible to the cardiotoxic effect of doxorubicin than human beings.

This study showed an alteration of the heart rate in dogs that received doxorubicin. Furthermore, episodes of sinus tachycardia were observed in all animals, similarly to previous descriptions in humans (Lefrak, 1973; Ali et al., 1979) and dogs (Susaneck, 1983; Crystal and Rush, 1991; Mauldin, 1992). The values of minimum and mean heart rate increased 44.6% and 41.7%, respectively, when compared to the means of control dogs. This finding is in agreement with Kehoe et al. (1978), and is possibly related to a compensatory effect to maintain cardiac function or to the high serum levels of catecholamines that can arise after doxorubicin administration. Dogs from group B also had an increase of 43.2% in the number of QRS complexes in comparison with animals from group A, which indicates an elevation of the heart rate in these dogs.

In this research, chronic administration of doxorubicin (180-240mg/m$^2$) led to the development of some type of arrhythmia in all the dogs from group B (5/5), being more frequent the ectopic supraventricular beats, which contributed to 93.7% of the total, whereas the ectopic ventricular beats represented 6.3% of the total. In a study by Kehoe et al. (1978), ectopic supraventricular beats were less frequent

and ectopic ventricular beats had a greater importance. Only number 4 dog showed a greater frequency of ectopic ventricular beats (51/59) in comparison with ectopic supraventricular beats (8/59). Supraventricular arrhythmias are likely to occur when high doses of doxorubicin are used. In some cases, however, supraventricular arrhythmias may precede ventricular ectopic beats. Since transient supraventricular arrhythmias may occur, as opposed by ventricular arrhythmias, it could eventually have recorded the 24-hour Holter when such arrhythmias happened in a greater frequency (Kehoe et al., 1978).

The presence of such arrhythmias was very important and significant, since it allowed to evaluate the degree of myocardial compromise due to doxorubicin cardiotoxicity, as described by Susaneck (1983), Cowan and Jacobs (1990), Crystal and Rush (1991), Mauldin et al. (1992), Jacobs (1996), Kittleson (1998) and Silva (2001).

In relation to the types of supraventricular arrhythmias, it was observed that all dogs from group B showed isolated atrial premature beats and amongst them, four (80%) also developed couplets of atrial premature beats and atrial tachycardia. In general, ectopic ventricular beats tend to increase its rate and become multiform with increasing duration of doxorubicin treatment (Kehoe et al., 1978). All animals from group B developed ventricular premature complexes sole or in couplets. Amongst them, three animals (60%) presented ventricular tachycardia and a single one (20%) developed bigeminism.

The evaluation of arrhythmias induced by the chronic treatment with doxorubicin indicates that this drug can exert a multiple action on cardiac electrophysiology, which characterizes the development of these several types of arrhythmias (Kehoe et al., 1978).

The 24-hour ambulatorial electrocardiographic evaluation also determined the occurrence of asystole or pauses greater than 2 seconds. Such abnormalities were detected in three animals (60%) from group B and in two animals (40%) from group A. The asystole periods were characterized by sinus arrest rather than caused by a significant alteration due to the cardiotoxic effect of doxorubicin. However, a physiologic effect may be related since this arrhythmia was especially observed when the animals were asleep, when an elevation of the vagal tone can occur, leading to an increase in parasympathetic action to the heart (Tilley, 1992). These data are similar to the findings described by Hall et al. (1991), who observed a frequent occurrence of sinus arrest ranging from 2.0 to 5.7 seconds in healthy dogs.

In this study, no ST segment shifts were observed in animals from group B. This finding is likely to be related to individual sensitivity to the cumulative dose of doxorubicin and the intoxication time in the animals of this group The 24-hour electrocardiogram was only performed 21 days after the last doxorubicin infusion. Although myocardial dysfunction was observed at that time, a longer observation did not show alterations in the ventricular repolarization phase suggesting a progression of the cardiotoxicity. Such findings were also reported in prior studies by Kehoe et al. (1978) and Souza (2004). On the contrary, Gralla et al. (1979), Mauldin et al. (1992) and Silva (2001) observed that ST depression is one of the main electrocardiographic signs in dogs submitted to a doxorubicin chronic treatment.

## CONCLUSIONS

The results sustain the hypothesis that ambulatorial electrocardiography (Holter system) is efficient in early determination of heart rate and cardiac rhythm alterations in dogs showing doxorubicin-induced myocardial dysfunction. Moreover, it was determined augmentations in minimum and mean heart rate, as well as the presence of cardiac arrhythmias, being supraventricular ectopies the most commonly recorded.

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