Species Differences in the Changes in Heart Rate and T-wave Amplitude after Autonomic Blockade in Thoroughbred Horses, Ponies, Cows, Pigs, Goats and Chickens

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ABSTRACT. Using Thoroughbred horses, ponies, Holstein cows, pigs, goats and chickens, the effects of atropine and/or propranolol on heart rate (HR) and T-wave amplitude (TWA) on ECG were examined. HR increased significantly in all animal species after the administration of atropine, and decreased in the pig, goat and chicken after the administration of propranolol. Parasympathetic predominance, estimated from the changes in HR induced by the autonomic blockers, was in the order of Thoroughbred horse > pony > cow > goat whereas the sympathetic predominance was in the order of chicken > pig. The positive ingredient in TWA was increased significantly in the Thoroughbred horse, pony, pig, and goat by atropine whereas decreased only in the pig by propranolol. —KEY WORDS: autonomic blockade, autonomic tone, heart rate, species difference, T-wave amplitude.

In previous studies, we have suggested that an alteration of the heart rate (HR) in domestic animals such as horses and cows through fetal, newborn and young stages might be closely related to the development of autonomic nervous function [14, 17] and that the changes in T-wave amplitude (TWA) in horses with growth [14] and by twitch restraint [16] might be due to the changes in autonomic nervous tone.

In recent years TWA has been investigated from neurological, ethological and psychological standpoints in human [3, 9, 19], horse [10, 13], pig [1, 4] and cow [5]. These studies indicate that TWA is influenced by autonomic modulators [4, 5], exercise or excitement [1, 10] and psychological factors [3, 9, 13, 19]. However, little is known on the relationship between TWA and autonomic nervous tone.

In the present study, the responses of HR and TWA to autonomic blockade were investigated using adult Thoroughbred horses, ponies, cows, pigs, goats and chickens, to elucidate the relationship between these responses and autonomic nervous tone.

MATERIALS AND METHODS

Animals and ECG recordings: The adult female animals used in the present investigation are listed in Table 1. These animals were reared under ordinary conditions in the Animal Husbandry Experiment Station of the University of Tokyo. Electrocardiograms (ECG) were recorded in A-B lead, a bipolar lead along the longitudinal heart axis or a base apex lead. Positive electrode was placed at the apex and the negative electrode at the base of the heart. This method has been standardized as a common bipolar lead in Japan. ECGs were recorded by an electrocardiograph at paper speeds of 25mm/sec for horses, cows, pigs and goats and by a pen recorder equipped with a biomedical amplifier at paper speeds of 50 or 100mm/sec for chickens. Needle elec-
trodes were used for cows, goats and chickens, and adhesive electrodes (Red Dot, 3M) for horses and pigs.

**Drugs and procedures:** ECG recordings were carried out under unaesthetized, unrestrained conditions in the stable (horses and goats), stanchion (cows) or stall (pigs). ECG recording in the chicken was done under unaesthetized, light restraint condition tying the wings and legs in the cage. Pigs were sometimes given feed during the experiment. ECGs were recorded for 3 min before and 10 min after the administration of the autonomic blockers.

Selective blockade of parasympathetic (muscarinic) and sympathetic (beta-adrenergic) nerves was accomplished with atropine sulphate (Sigma Chemicals) and propranolol (Sigma Chemicals), respectively. Drugs were injected into the jugular vein (horses, cows and goats), ear vein (pigs) or wing vein (chickens). A single dose of atropine and propranolol, and a combined dose of both drugs were given as detailed in Table 1. HR after the combined administration was taken as the intrinsic heart rate without parasympathetic and sympathetic neural regulation, as indicated by Jose [11].

The drugs were tested in random order. At least one-day interval was provided between two successive tests. The dosage was determined by preliminary studies and also by referring to previous reports on the horse [6], cow [7], pig [4, 12], sheep [8, 21, 22, 24] and chicken [20].

**HR, TWA and autonomic nervous tone measurements and statistics:** HR (beats/min) before the administration of an autonomic blocker was calculated by averaging the mean RR interval every 10 sec for 3 min before the administration, while HR after autonomic blockade was calculated by averaging the mean RR interval for 10 sec at 3, 4, 5, 6 and 7 min after the administration. TWA was measured on several arbitrarily selected T-waves in the 10-sec recordings used for HR measurements. In order to compare the relative effect of both components of the autonomic nervous system, the following equations proposed by Walsh [23] and Sturkie *et al.* [20] were employed:

\[
\text{Sympathetic tone (\%) = } \frac{(HR_{at}-HR_{at+pr})}{HR_{at+pr}} \times 100
\]

\[
\text{Parasympathetic tone (\%) = } \frac{(HR_{pr}-HR_{at+pr})}{HR_{at+pr}} \times 100
\]

\[
\text{Net autonomic tone (\%) = } \frac{(HR_{at}-HR_{at+pr})+(HR_{pr}-HR_{at+pr})}{HR_{at+pr}} \times 100
\]
SPECIES DIFFERENCE IN HEART RATE AND T-WAVE

<table>
<thead>
<tr>
<th></th>
<th>Atropine</th>
<th>Propranolol</th>
<th>Atropine plus propranolol</th>
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<tbody>
<tr>
<td></td>
<td>Before</td>
<td>After</td>
<td>Before</td>
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<tr>
<td>Thoroughbred 5</td>
<td>28.2±1.3a)</td>
<td>81.4±6.2**b)</td>
<td>33.0± 0.9</td>
</tr>
<tr>
<td>Pony n=6</td>
<td>45.6±1.1</td>
<td>97.4±5.3**</td>
<td>44.8± 1.7</td>
</tr>
<tr>
<td>Cow n=6</td>
<td>64.6±4.1</td>
<td>73.0±1.7**</td>
<td>61.0± 5.1</td>
</tr>
<tr>
<td>Pig n=7</td>
<td>109.0±5.9</td>
<td>158.0±8.9*</td>
<td>104.8± 4.1</td>
</tr>
<tr>
<td>Goat n=6</td>
<td>83.2±4.1</td>
<td>130.8±3.9**</td>
<td>85.6± 5.3</td>
</tr>
<tr>
<td>Chicken n=6</td>
<td>329.0±9.6</td>
<td>365.0±7.7**</td>
<td>354.0±14.7</td>
</tr>
</tbody>
</table>

a) Mean±SE (beats per min).

where HR.at, HR.pr and HR.at+pr are HR after the administration of atropine, propranolol, and atropine plus propranolol, respectively. The response to each treatment was analyzed by Student's paired t-test. Differences between means were considered to be statistically significant when the p value was less than 0.05. The results are expressed as means or mean±SE in the text.

RESULTS

The results obtained for HR are summarized in Table 2. The average HR before the administration of autonomic blockers (resting heart rate, beats/min) was 30.0 in the Thoroughbred horse, 44.1 in the pony, 62.1 in the cow, 109.5 in the pig, 85.1 in the goat, and 346.7 in the chicken. The percent change in HR in response to parasympathetic blockade was +190.4±23.5% in the Thoroughbred horse, +113.1±8.2% in the pony, +59.4±9.6% in the goat, +14.5±4.9% in the cow, +43.0±4.7% in the pig and +10.8±1.4% in the chicken. The percent change in HR induced by sympathetic blockade was −30.4±2.1% in the chicken, −14.0±1.8% in the pig, −12.3±1.9% in the goat, −5.35±1.7% in the cow, −0.48±1.0% in the pony and +1.36±1.9% in the Thoroughbred horse. The percent change in HR due to both parasympathetic and sympathetic blockades was −29.2±2.4% in the chicken, −3.43±4.3% in the pig, +24.3±9.7% in the cow, +33.1±13.2% in the goat, +127.2±19.8% in the pony and +190.3±20.4% in the Thoroughbred horse.

As shown in Fig. 1, the sympathetic tone was greater in the chicken, pig and goat than in the cow, pony and Thoroughbred horse. On the other hand, the parasympathetic tone ranked in order of Thoroughbred horse > pony > goat > cow > pig > chicken. From the calculation of the net autonomic tone, it was shown that the chicken and pig were of the sympathetic-predominant type, and the Thoroughbred horse, pony, cow, and goat were parasympathetic-predomi-
Fig. 2-A. ECGs before and after the administration of atropine (A), propranolol (B) and atropine plus propranolol (C) in Thoroughbred horse, pony and cow (A-B lead).

Fig. 2-B. ECGs before and after the administration of atropine (A), propranolol (B) and atropine plus propranolol (C) in pig, goat and chicken (A-B lead).

nant.

Fig. 2 (A and B) shows typical ECG recordings before and after the administration of the autonomic blockers in each animal. Changes in TWA are shown in Figs. 3-5. After the administration of atropine, the positive ingredient in TWA increased significantly in Thoroughbred horses, ponies, pigs and goats but not in cows and chickens. After the administration of prop-
SPECIES DIFFERENCE IN HEART RATE AND T-WAVE

Fig. 3. Changes in the T-wave amplitudes after the administration of atropine (mean±SE). Asterisks indicate significant differences between the heart rates before and after the administration of atropine. P<0.05 (*), P<0.01 (**).

Fig. 4. Changes in the T-wave amplitudes after the administration of propranolol (mean±SE). Asterisks indicate significant differences between the heart rates before and after the administration of propranolol. P<0.05 (*).

Fig. 5. Changes in the T-wave amplitudes after the administration of atropine plus propranolol (mean±SE). Asterisks indicate significant differences between the heart rates before and after the administration of atropine plus propranolol. P<0.05 (*), P<0.01(**).

ranolol, the positive ingredient in TWA decreased significantly only in pigs. On the other hand, after the combined administration of atropine and propranolol, the positive ingredient significantly increased in the Thoroughbred horses and ponies, significantly decreased in pigs, and did not change in cows, goats and chickens.

DISCUSSION

Jose [11] has shown that simultaneous application of beta-adrenergic blockade, propranolol, and cholinergic blockade, atropine, can produce functional denervation in human heart, during which intrinsic cardiac function, particularly, intrinsic heart rate, can be assessed. Using the same procedure, autonomic control of the resting heart rate has been studied in horses [6], cows [7], pigs [4, 12], sheep [8, 21, 22, 24] and chickens [20]. In the present experiment, we used this method to estimate the relationship between TWA and autonomic nervous tone in several animal species.

The results indicate that the resting heart rate in each animal is modulated by variations in sympathetic and parasympathetic tone, which show considerable species-dependent differences (Fig. 1). The species ranking for the net autonomic nervous tone (parasympathetic predominance < sympathetic predominance) was Thoroughbred horse < pony < cow < goat < pig < chicken. The intrinsic heart rate in these animals, except for the chicken, ranged from 74 to 110 beats per min, while their resting heart rate ranged from 30.0 to 109.5 beats per min. These results suggest that the resting heart rate in these animals is kept lower than their intrinsic heart rate by the parasympathetic activity.

The polarity of the T-wave depends essentially on the lead method for ECG recording. However, it also varies according to the animal species even if the same lead
method is used. In fact, the configuration and amplitude of the T-wave in the A-B lead differ among the Thoroughbred horse, pony, cow, pig, goat and chicken, as shown in Fig. 2.

In horses, the form of T-wave changes markedly with growth [14], twitch restraint [16], human contact [13] and exercise [10]. An increase in the positive ingredient of TWA due to exercise or human contact is accompanied by an increase in HR and an increase in the negative ingredient due to twitch restraint is accompanied by a decrease in HR. In the present study, the positive ingredient of TWA in Thoroughbred horses and ponies was markedly increased by atropine. The increased TWA as well as HR are probably resulted from sympathetic predominance following parasympathetic blockade. The present result supports the suggestion made by Fregin [2] that functional variations in autonomic nervous tone alters TWA in horses even in the absence of clinical evidence of disease.

Gregory et al. [4] reported that TWA in stress-sensitive pigs is influenced by alpha-adrenergic activity in the absence of beta-adrenergic and parasympathetic receptor activation. Faraci et al. [1] reported that prominent increases in TWA of the pig induced by exercise result from hyperkalemia. In the present study using normal adult pigs, the positive ingredient of TWA was significantly increased by atropine and decreased significantly by propranolol and by atropine plus propranolol. These results could not be compared directly with the above-mentioned finding of Gregory et al. [4], because they used stress-sensitive pigs. However, it is considered that the change in TWA in the pig might be closely related to the autonomic nervous tone in either case.

In the present study, the positive ingredient of TWA was significantly increased by atropine in the Thoroughbred horse, pony, pig and goat. This increase may be attribut-
要約
自律神経遮断薬投与後の心拍数、心電図T波の変化にみられる動物種差：松井寛二・菅野 茂
（東京大学農学部附属牧場、家畜環境生理学教室）—ウマ（サラブレッド種およびボニー）、ウシ（ホルスタイン種）、ブタ、ヤギ（シバヤギ）およびニワトリの安静時にアトロピン、プロプラノロールの単独ならびに同時投与を行い、心拍数、心電図T波に現われる反応を調べた。動物種により程度の差はあるが、アトロピン投与により心拍数は有意に増加し、プロプラノロール投与ではブタ、ヤギ、ニワトリでのみ減少した。自律神経遮断薬投与後の心拍数から算出した自律神経緊張度はサラブレッド＞ボニー＞ウシ＞ヤギの順に副交感神経緊張型を示し、また、ニワトリ＞ブタの順に交感神経緊張型を示した。T波の電位はアトロピン投与によりサラブレッド、ボニー、ブタ、ヤギにおいて陽性成分が有意に増大し、プロプラノロール投与では、ブタのみで減少した。このようなT波の電位変化には自律神経緊張度が密接に関係し、動物種により明らかな差があることが示唆された。