Tr. J. of Veterinary and Animal Sciences
22 (1998) 421-426

Effects of Different Concentrations of Monensin on the Electrocardiogram and the Serum Ion Balance of the Rabbit

İsmail MERAL, Ali ÇINAR, Fahri BAYIROĞLU
Yüzüncü Yıl University, Veterinary Collage, Department of Physiology, Van-TURKEY

S. Oktay ASLAN
Harran University, Medical School, Department of Pharmacology, Şanlıurfa-TURKEY

Received: 16.01.1997

Abstract: The present study was planned to investigate the effects of two different concentrations (10 and 40 mg/kg) of monensin, carboxylic ionophore antibiotic, on the ECG and the serum Na^+ and K^+ concentrations of rabbits. Fifteen male rabbits, about 2 kg and 1 year old, were used. ECGs were recorded by a direct writing electrocardiograph. Blood samples for measuring serum Na^+ and the K^+ concentrations were collected and the leads I, II, II, aVR, aVL and aVF were recorded before and 6 hours after monensin was given. The durations and amplitudes of waves on the trace were measured in lead II and electrical axis also measured in leads I and III. We found that the effects of monensin on ECG and the serum Na^+ and the K^+ concentrations were dose dependent. In low doses (10 mg/kg) it produced a sinusoidal tachycardia by decreasing the time interval between two impulse production by sinoatrial node without changing the serum ion balance. However, in high doses (40 mg/kg) it produced a sinusoidal tachycardia with increasing the serum K^+ and decreasing the serum Na^+ concentrations.

Key Words: Monensin, electrocardiogram, serum, sodium, potassium, rabbit

Monensinin Farklı Dozlarının Tavşanlarda Elektrokardiogram ve Serum İyon Dengesi Üzerine Etkisi

Özet: Bu çalışma bir karboksilik antibiyotik olan monensinin iki farklı dozunun (10 ve 40 mg/kg) tavşan EKG ve serum Na^+ ve K^+ konsantrasyonu üzerindeki etkisini araştırmak amacıyla yapılmıştır. Çalışmada yaklaşık 2 kg ağırlığında ve 1 yaşında 15 tavşan kullanıldı. Monensin uygulamasından önce ve 6 saat sonra serum Na^+ ve K^+ konsantrasyonunu ölçmek için kan alındı ve I, II, III, aVR, aVL ve aVF derivasyonları kaydedildi. Dalgaların boyu ve süreleri II. derivasyonda hesaplandı. Ayrıca I. ve III. derivasyonlar kullanılarak normal elektriksel eksen hesaplandı. Monensinin EKG ve serum Na^+ ve K^+ konsantrasyonu üzerindeki etkisinin doza bağımlı olduğu bulundu. Düşük dozda (10 mg/kg) monensin sinuzoidal taşikardi oluşturduğu halde serum Na^+ ve K^+ konsantrasyonunu değiştirmedi. Ancak daha yüksek dozda (40 mg/kg) monensin hem sinuzoidal taşikardi oluşturdu ve hem de serum K^+ konsantrasyonunu artırarak serum Na^+ konsantrasyonunu düşürdü.

Anahtar Sözcükler: Monensin, elektrokardiogram, serum, sodyum, potasyum, tavşan

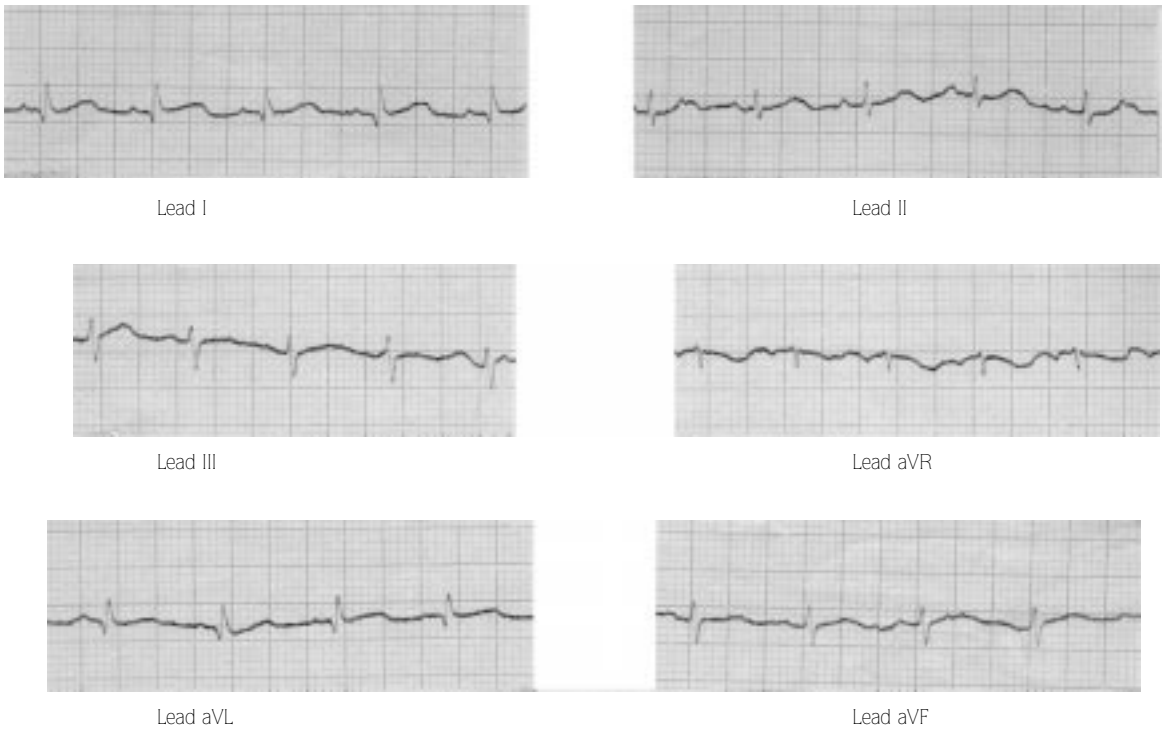
Introduction

The electrocardiogram (ECG), record of the potential fluctuations during a cardiac cycle, gives informations about all details of cardiac activity (1) and is recognized as a laboratory aid in diagnosis cardiovascular diseases (2, 3). Many studies have showed (4, 5) that some diseases, environmental conditions, breed, age and muscle condition can affect the ECG.

Monensin is a carboxylic ionophore antibiotic which is derived from the fermentation of the fungus *Streptomyces cinnemonensis* (6). It is used as a coccidiostat in birds, lambs and calves (7, 8, 9) and primarily as a growth promoter (10, 11) and as a bloat preventive (8, 12) in cattle.

Monensin has also a strong positive inotropic effect in cardiac muscle (13, 14, 15). This effect occurs, because as an ionophore, monensin has strong affinity for $(Na^+)_o$. The affinity of monensin for Na^+ is ten times that for K^+, its nearest competitor in biological systems (16). The monensin-induced increase in $(Na^+)_i$ facilitates the entry of Ca^{2+} into the cell by a Na^+ (out) / Ca^{2+} (in) exchange mechanism (17). This Ca^{2+} shift is the primary factor mediating the cellular response. Another factor modifying the cellular response includes the alteration of the pH of intracellular components (pH_i) since monensin increases the pH_i by transferring H^+ out of the cell (16.)

Figure 1. The leads of ECGs of the control rabbits (standardization, 1mV=10mm; chart speed, 50 mm/sec.



Meral et al. demonstrated that monensin increases the contraction force of the papillary muscles isolated from the guinea-pigs (17) by increasing the intracellular Ca^{2+} concentration. He also suggested that monensin should be studied for the treatment of congestive heart failure (17). However, it has also become clear that, in addition to increasing the force of contraction, increased intracellular Ca^{2+} concentration has other effects on cardiac muscle. In particular, increases in the intracellular Ca^{2+} concentration appear to initiate some cardiac arrhytmias. Meral et al. also demonstrated by using single myocytes that monensin increases the intracellular Ca^{2+} concentration by increasing the intracellular Na^{+} concentration (17). Since monensin increases the intracellular Na^{+} concentration, it should also decrease the serum Na^{+} concentration and increase the serum K^{+} concentration. The present study was planned to investigate the effects of two different concentrations of monensin on the ECG and the serum Na^{+} and K^{+} concentrations of rabbits.

Material and Methods

Preparation of animals for recording:

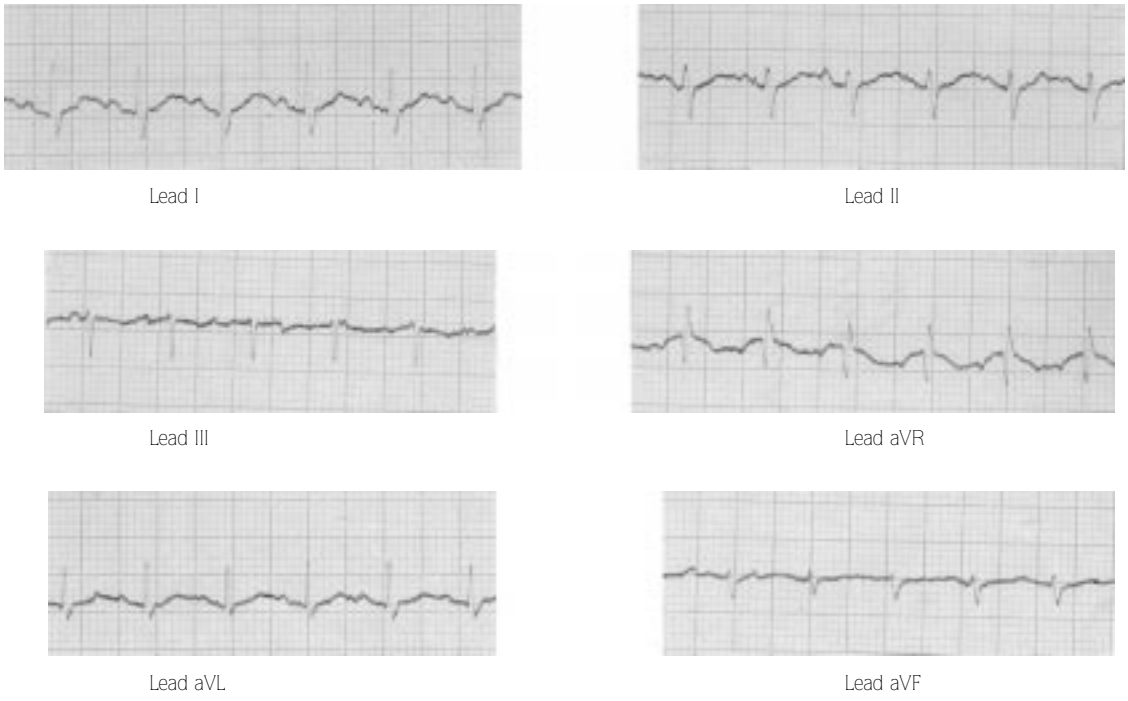
Fifteen male rabbits, about 2 kg and 1 year old, were used. Alligator clip electrodes were attached to the skin at the triceps brachii muscle (coput longum and coput laterale) of the right and left limbs and biceps femoris muscle of the right and left hips. Electrode gel was rubbed into the skin in the area where the alligator clips were attached to act as a decreasing agent and thereby decrease the resistance of the skin (18). The rabbits were immobilized by wrapping a light cotton around them and then placed on a table. We waited about 10 min. for rabbits to get calm. The rabbits were not anesthetized at any time. All recordings were made on the same day.

ECGs were recorded by a direct writing electrocardiograph (Cardiofax 6851; Nihon Kohden, Tokyo). All ECGs were standardized at 1 mV = 10 mm, with a chart speed of 50 mm/sec. Leads I, II, II, aVR, aVL and aVF were recorded before and 6 hours after monensin was given. The durations and amplitudes of waves on the trace were measured in lead II and electrical axis also measured in leads I and III.

Measurement of the serum Na^{+} and K^{+} concentrations:

Blood samples for measuring serum Na^{+} and the K^{+} concentrations were collected before and 6 hours after

Figure 2. The leads of ECGs of the 10 mg/kg monensin treated groups (standardization, 1 mV=10mm; chart speed, 50 mm/sec.)



Lead I Lead II

Lead III Lead aVR

Lead aVL Lead aVF

monensin was given. The Na^+ and the K^+ concentrations were measured by using a $Na^+/K^+/C^{1-}$ analyzer (IL test™ Solution Pact-ILyte $Na^+/K^+/Cl^-$, USA).

Drug used:

Two different concentrations (10 and 40 mg/kg) of monensin (Kartal Kimya San. ve Tic. A. Şti, İstanbul, Türkiye) were used. Monensin was dissolved in distillated water and given directly into the stomach by using a sterile plastic catheter.

Statistical analysis:

Three treatments (control, 10 mg/kg and 40 mg/kg monensin) were used. Each treatment contained same number (n=5) of rabbits with same sex and weight. The data were expressed as mean ± standard deviation (SD) and analyzed using analysis of variance (ANOVA). Least significant difference (24) was used to test for differences among means for which ANOVA indicated a significant ($P \leq 0.05$) F ratio.

Results

Figure 1,2 and 3 provide all leads of the ECG of the normal and monensin treated rabbits respectively. These readings are representative of the majority of rabbits. The durations and amplitudes of all waves in lead II and the serum Na^+ and K^+ concentrations are shown in Table 1.

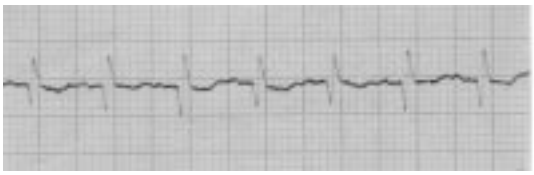
In control group, the P wave was negative in lead aVR and positive in other leads. The mean duration of the P wave was 0.020 sec (0.016-0.025 sec) and its mean average amplitude was 0.10 mV (0.08-0.13 mV). Q wave was only seen in leads I and aVL. The mean duration of QRS complex was 0.040 sec (0.035-0.045 sec) and its mean amplitude was 0.50 mV (0.40-0.63 mV). T wave, like Q wave, was negative in lead aVR and positive in other leads. The mean duration of the T wave was 0.080 sec (0.05-1.12 sec) and its average amplitude was 0.25 mV (0.19-0.28 mV). The average value of the mean electrical axis of the rabbit heart was + 65° (-20° to + 130°). The heart rate of rabbits was 187.6 beats/min. (158-222 beats/min.).

Ten and 40 mg/kg monensin treatments did not change the durations or amplitudes of any wave on the trace except they decreased ($P<0.05$) the average amplitude of T wave (from 0.25 mV to 0.15 and 0.10 mV respectively). They also increased ($P<0.05$) the heart rate (from 187.6 beats/min. to 300 and 330 beats/min. respectively) by producing a P+T wave and decreasing the time interval between two impulse production in sinoatrial node (sinusoidal tachycardia).

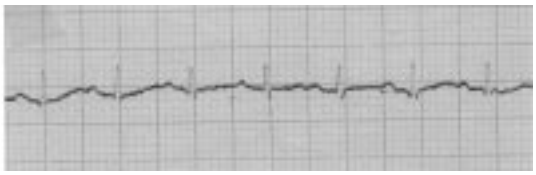
Variables	Control	10 mg/kg monensin treated	40mg/kg monensin treated
P(sec)	0.020±0.005	0.030±0.005	0.020±0.004
P(mV)	0.10±0.02	0.10±0.02	0.100±0.02
QRS(sec)	0.040±0.006	0.040±0.005	0.030±0.005
QRS (mV)	0.50±0.11	0.60±0.14	0.5+0.09
T (sec)	0.080±0.022	0.10±0.033	0.10±0.029
T(mV)	0.25±0.05	0.015±0.03*	0.10±0.03*
P-Q (sec)	0.70±0.018	0.060±0.015	0.080±0.0.019
Q-T (sec)	0.18±0.03	0.19±0.03	0.17±0.03
Heart rate	1876±29	300±37*	330±43*
Mean electrical axis	+65° (-20° to + 135°)	+65° (-20° to + 135°)	+65° (-20° to + 135°)
Serum Na^+ concentration	138.7±4.3	143.4±56	111.8±5.2*
Serum K^+ concentration	5.3±0.8	4.9±0.8	61.37±6.5*

Table 1. Amplitudes and durations of waves, heart rates and the serum Na^+ and K^+ concentrations of control, 10 mg/kg monensin treated and 40 mg/kg monensin treated rabbits (mean±standard deviations are shown, n=15). *denotes a significant difference between control and the treatment group.

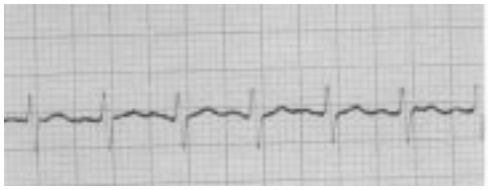
Figure 3. The leads of ECGs of the 40 mg/kg monensin treated groups (standardization, 1mV=10mm; chart speed, 50 mm/sec).



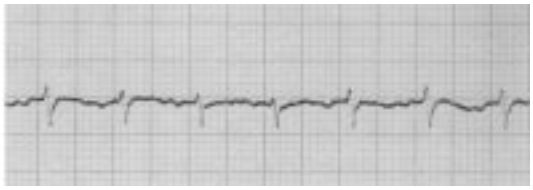
Lead I



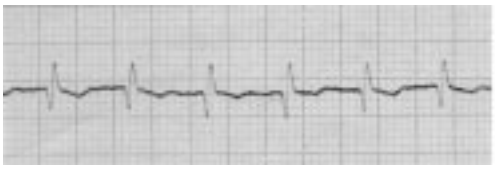
Lead II



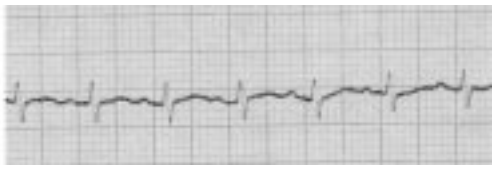
Lead III



Lead aVR



Lead aVL



Lead aVF

Although 10 mg/kg monensin treatment did not change the serum Na^+ or K^+ concentrations, 40 mg/kg monensin significantly ($P<0.05$) decreased the serum Na^+ concentration (from 138.7±4.3 to 111.8 ± 5.2) and increased the serum K^+ concentration (from 5.3 ± 0.8 to 61.37 ± 6.5).

Discussion

This study was undertaken to evaluate the toxic effect of the different concentrations of monensin on the ECG and the serum Na^+ and K^+ concentrations of rabbits. Data presented in this study showed that monensin treatment produced a sinusoidal tachycardia by decreasing the time interval between two impulse

production by sinoatrial node. It has been suggested (19) that the effect of monensin on systemic blood pressure and the heart rate is dose-dependent; in low doses there appear to be a slight but statistically insignificant decrease in aortic pressure and a slight increase in the heart rate. Sani et al (19) demonstrated that heart rate had a tendency to increase by a few beats perminute during the first 5 minute after the 200 µg/kg of monensin, but essentially remained unchanged.

In the present study we found that 10 and 40 mg/kg monensin increased the heart rate significantly ($P<0.05$). Dose of monensin in this experiment was high (toxic). It has been suggested that (19) monensin releases both norepinephrine from the adrenergic nerve terminals and epinephrine from the adrenal medulla, since the cardiovascular actions of the large dose of monensin are similar to the those reported for both norepinephrine and epinephrine. The Ca^{2+} influx is an essential step in catecholamine release from adrenal medulla, chromoffin granules and adrenergic nerve terminals (20, 21). How monensin can cause an increased influx of Ca^{2+} remains unexplained at the present time. However, experiments in isolated rabbit atria (22, 23) suggest that monensin does not directly transport Ca^{2+} across the cell membrane, but may do so indirectly via the transport of Na^{+}, which then exchanges for Ca^{2+} by a Na^{+} (out)/ Ca^{2+} (in) exchanger (17). The Ca^{2+} that enters the cell (by a Na^{+} (out)/Ca^{2+} (in) exchanger and by a catecholamine release) causes an additional release of Ca^{2+} from the sarcoplasmic reticulum (Ca^{2+}-induced Ca^{2+}-release). Release of catecholamines from adrenal medulla, chromoffin granules and adrenergic nerve terminals by monensin causes an increase in the impulse production in sinoatrial node causing a sinusoidal tachycardia. In our previous experiments we found that monensin caused a positive inotropic effect in guinea-pig papillary muscles (17). Other positive inotropic drugs, such as digoxin also causes a positive inotropic effect by increasing the intracellular Ca^{2+} concentration but it also produces a ventricular tachycardia (17). However, in this experiment monensin did not produce any serious arhytmia except sinusoidal tachycardia that can be easily treated by using parasymphatic drugs. This finding may be important for the use of monensin in the treatment of the congestive heart failure that we suggested before (17). However, more studies are needed for the use of monensin the treatment of the congestive heart failure.

It has been also found that the average value of the mean electrical axis of the rabbit heart was + 65° (-20° to +130°). This result indicated that the ventricular axis is from right to left and from front to back in rabbits.

We also found that 10 mg/kg monensin treatment did not change the serum Na^{+} or K^{+} concentrations. This result indicated that 10 mg/kg monensin is not toxic enough to produce any change in the serum ion balance although it is toxic enogh to produce an increase in the heart rate. However, 40 mg/kg monensin significantly decreased the serum Na^{+} concentration and increased the serum K^{+} concentration. This indicated that in high (very toxic) doses monensin, in addition to the increased heart rate, also changes the serum ion balance by transferring Na^{+} into the cells (24, 25) and causing an increased intracellular Na^{+} concentration and decreased serum Na^{+} concentration.

We concluded that the effects of monensin on ECG and the serum Na^{+} and the K^{+} concentrations are dose dependent. In low doses it increases the heart rate without changing the serum ion balance. However, in high doses it increases the heart rate with a changed serum ion balance.

Acknowledgments

We would like to express our sincere gratitude to Dr. Frederick B. Hembrough, Veterinary Medicine, Iowa State University, Ames, IA, the USA for his help in various ways during our work. We would also like to thank Kartal Kimya San ve Tic. A. Şti, İstanbul, Türkiye for providing us the monensin we used in our experiment.

References

1. Bopelet, M.: Normal electrocardiogram of the chicken: its variation during vagal stimulation and following vagotomies. Comp. Biochem. Physiol. 1974; 47A, 361-369.
2. Baulianne, M., Hunter, D.B., Julian, M., O'Grady, R. and Physick Sheard, P.W.: Cardiac muscle mass distribution in the domestic Turkey and relationship to electrocardiogram. Avian Dis. 1992; 36, 582-589.
3. Sturki, P.D.: Effects of changes in position the heart of the chicken on the electrocardiogram. Am. J. Physiol. 1948; 154, 251-257.
4. Czarneckie, C.M. and Good, A.L.: Electrocardiographic technique for identifying developing cardiomyopathies in young turkey poults. Poult. Sci. 1980; 59, 1515-1520.
5. Emre, B., Sulu, N., Bağcı, C., Pişkin, I. and Çınar, A.: Denizli horozlarında elektrokardiogram. Fac. Vet. Med. Univ. Ankara. 1993; 40,4, 543-551.

6. Jr. Haney, M.E. and Hoehn, M.M.: Monensin, a new biologically active compound: I. Discovery and isolations. Antimicrob. Agents Chemother. 1967; 349-352.

7. Fahim, M. and Pressman, B.: Cardiovascular effects and pharmacokinetics of the carboxylic ionophore monensin in dogs and rabbits. Life Sci. 1981; 29, 1959-1966.

8. Van Vleet, J.F. and Ferrans, V.J.: Ultrasonic alterations in skeletal muscle of pigs with acute monensin myotoxicosis. Am. J. Pathol. 1984; 114, 3, 461-471.

9. Elsasser, T.H.: Potential interactions of ionophore drugs with divalent cations and their function in the animal body. J. Anim. Sci. 1984; 59, 3, 845-853.

10. Thornton, J.H., Ovens, F.N., Lemenager, R.P. and Tatusek, R.: Monensin and ruminant methane production. J. Anim. Sci. 1976; 43, 336 (Abstr).

11. Potter, E.L., Cooley, C.O., Richard, L.F., Roun, A.P. and Rathmacher, R.P.; Effect of monensin on performance of cattle fed forage. J. Anim. Sci. 1976; 43, 665-669.

12. Mitema, E.S. and Sangiah S.: Effects of some calcium modulators on monensin toxicity. Vet. Hum. Toxicol. 1988; 30,5, 409-413.

13. Mollennauer, H.H., Morre, D.J. and Rowe, L.D.: Alteration of intracellular traffic by monensin; mechanism, specificity and relationship to toxicity. Biochim. Biophys. Acta. 1990; 1031, 225-246.

14. Pressman, B.C.: Biological applications of ionophores. Ann. Rev. Biochem. 1976; 45, 501-529.

15. Sutko, J.L., Besch, H.R., Bailey, J.C., Zimmerman, G. and Watanabe, A.M.: Direct effects of the monovalent cation ionophores monensin and nigericin on myocardium. J. Pharmacol. Exp. Ther. 1977; 203, 685-700.

16. Shlafer, M. and Kane, P.: Subcellular actions and potential adverse cardiac effects of the cardiotonic ionophore monensin. J. Pharmacol. Exp. Ther. 1980; 214, 567-573.

17. Meral, I.: Utilization of fatigued and non-fatigued papillary muscles and ventricular myocytes on the comparison of inotropic, chronotropic and intracellular calcium changes induced by monensin and digoxin. 1996; Iowa State University, Veterinary Collage, the USA (Doktora tezi).

18. Sulu, N., Çınar, D.A. and Özdemir, S.: Verapamil HCl'nin tavuklarda elektrokardiogram üzerine etkisi. Fac. Vet. Med. Univ. Ankara. 1993; 40, 2, 281-291.

19. Saini, R.K., Hester, R.K. and Pressman, B.C.: Characterization of the coronary vasodilator and hemodynamic actions of monensin, a carboxylic ionophore. J. Cardiovas. Pharmacol. 1979; 1, 123-138.

20. Douglas, W.W. and Rubin, R.P.: The role of calcium in the secretory response of the adrenal medulla to acetylcholine. J. Physiol (Lond). 1961; 158, 40-57.

21. Burn, J.H. and Gibbson, W.R.: The release of noradrenaline from sympathetic fibers in relation to calcium concentrations. J. Physiol (Lond). 1966; 181, 214-223.

22. Shlafer, M., Somani, P., Pressman, B.C. and Palmer, R.F.: Effects of the carboxylic ionophore monensin on atrial contractility and Ca2+ regulation by isolated cardiac microsomes. J. Mol. Cell Cardiol. 1978; 10, 33-346.

23. Shlafer, M., Pressman, B.C., Somani, P. and Palmer, R.F.: Inotropic and chronotropic effects of monensin, a carboxylic ionophore, on isolated rabbit atria. Circulation 1975; 52, II, 42-51.

24. Snedecor, GW. and Cochran, WG.: Statistical methods. Eight edition. 1989; 227