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Heart performance of lambs and its relation to muscle volume and body surface

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Abstract: Electrocardiography-gated dynamic magnetic resonance imaging (MRI) methodology was developed for the in vivo examination of sheep heart characteristics. By combining noninvasive determination of skeletal muscle mass, we studied the relationship between total body skeletal muscle content and heart performance. Measurements were carried out on male Merino lambs using 1.5 T field strength equipment. Average left ventricular volumes were determined and stroke volume was estimated during image postprocessing. Ejection fraction was calculated (73 ± 1.8%) and the cardiac output (CO) value was estimated (2.75 ± 0.16 L/min). After measuring left ventricular wall thickness, contraction values were determined at the septum (62%), anterior (69%), lateral (54%), and posterior (58%) walls. Ventricular mass was also calculated. Body composition measurement was performed immediately after MRI examination by computerized tomography (CT) during the same narcosis. Relative CO value was developed from the interpretation of the functional MRI and volumetric CT results, expressing the relationship between heart performance and total body skeletal muscle volume. Finally, CO value relating to body surface was estimated (18.3 ± 3.1 $dm^2/(L\ min^{-1})$) to characterize the metabolic rate.

Key words: Lamb, magnetic resonance imaging, computerized tomography, cardiac performance

1. Introduction

The use of electromagnetic waves is gaining increasing importance in different areas of animal science, from meat quality determination (1,2) to in vivo evaluation of body composition, and is applied as a possible means for the genetic improvement of a wide variety of species (3–5).

Moreover, the use of animal models (mouse, rat, rabbit, canine, swine, and sheep) in human cardiac research has offered an invaluable opportunity to develop new diagnostic and therapeutic methods (6,7).

Magnetic resonance imaging (MRI) was first utilized as an in vivo technique for the determination of body composition of animals by Kallweit et al. (8). A study of sheep indicates that the quantification of lean and fat tissue by means of prediction equations is highly accurate (9). The description of electrocardiography (ECG)-gated MRI for the in vivo measurement of heart performance in pigs was presented by Petrási et al. (10), and an ovine medical model was first published by Pilla et al. (11). Recently, sheep have been used as a model for cardiac MRI of different heart failures, such as dilated cardiomyopathy, based on T1 mapping sequence (12). An ovine model measured end-systolic volume after myocardial infarction using the MRI method, with and without micropump-based partial mechanical circulatory support (13). The cardiac MRI method and computerized tomography (CT) were used jointly to measure the functional heart performance of the pig and heavy turkey in connection with body tissue composition determination (14,15). In addition, real-time 3D echocardiography, as applied by Schmidt et al., may be a useful quantitative method for the determination of ovine cardiac characteristics (16). Moreover, Segers et al. (17) developed an indirect prediction method for the characterization of heart performance in sheep.

The results achieved in intensive pigs and turkeys indicate disadvantageous changes in heart performance caused by continuous selection for the increase of skeletal muscle volume. In this respect, sheep seem to be less affected at present. However, this situation may change in the future, if selection for increased muscle building capacity continues.

The goal of this experiment was to develop an appropriate in vivo method for the quantitative measurement of sheep heart performance and to determine basic heart performance data jointly with skeletal muscle volume and body surface estimation.

2. Materials and methods

Methodological measurements were carried out on 3 male Merino lambs at a live weight of 20 kg (60 ± 4 days of age). Investigations were performed at the Health Center of Kaposvár University, using Siemens Magnetom

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Vision Plus magnetic resonance tomograph equipment of 1.5 T magnetic field strength and a Somatom S40 spiral CT scanner (Siemens, Germany). The lambs were premedicated intramuscularly with Rometar (xylazine 2%; Spofa; 0.2 mg/body weight kg). Following inhalation, anesthesia was introduced through a narcotic mask using 3 vol.% isoflurane (Abbott Laboratories, USA) until total relaxation. The animals were then intubated and attached to a narcotic unit using a Penlon evaporator and Ohmeda flowmeter. Continuous deep narcosis was obtained using 1.5–2 vol.% isoflurane and 2 vol.% oxygen as carrier gas, according to the recommendation of Hikasa et al. (18).

The MRI examination was conducted using ECG-gated sequences (19). A special active electrode (Bruker Medical) was used to obtain proper signal strength from the electrocardiograph. The electrode was attached to the signal cable at 10 cm from the sternum, on the left side, between ribs 3 and 6. The other 2 were directed to the left olecranon between ribs 3 and 6. During the imaging process, the animals were placed into a special MRI-compatible plastic container in ventral position with extended limbs, and were then fixed with belts. At first, quick images were taken to locate the heart according to the coordinate system of the body. Subsequently, localization images were taken to allocate the longitudinal axis of the heart in the sagittal, coronal, and transversal planes. Multislice-multiphase images were then taken orthogonally to the longitudinal axis of the heart, from the apex to the base, for the acquisition of prospective data (10). Total data acquisition lasted approximately 10–12 min. Overall, 9 slices and 9 images (phases) within each slice were acquired according to a single heart cycle (single slice-multiphase). The ventricles were covered by 6 of 9 transversal slices representing the total heart volume. The phases applied were characterized by the following data: echo time 6.8 ms, repetition time 60.0 ms, θ 30°, field of view 400–500 mm, matrix size 256 × 256 pixels, slice thickness 8 mm, and slice gap 1 mm. Moreover, the left ventricular volumes were measured and stroke volume (SV) was calculated as the difference between the end-diastolic and end-systolic data. The calculated ejection fraction (EF) is given as the percentage of the ratio of the SV to the end-diastolic volume. The left ventricular mass was calculated from the regions epicardium – endocardium + papillary 1 + papillary 2 (as marked in Figure 3) × 1.05 (g/cm^3). The cardiac output (CO) value was estimated as a product of the SV and heart rate. Wall thickness was measured by the septum and the anterior, lateral, and posterior walls, 9 mm beneath the atrioventricular valve. Images were evaluated using MASS 4.1 software.

Serial CT images were taken continuously from the neck (atlas) to the hock with 10-mm slice thickness. Total body skeletal muscle volume and total body surface were measured. The image analysis performed was described in detail by Romvári et al. (15).

3. Results

Heart rate was monitored before and during the MRI examination. Total pulse in relaxed state was registered at 89 ± 3 beats/min. The electrocardiogram attained from the foregoing time of examination gave accurate information about the overall conditions and the anesthetic possibilities concerning each animal.

The MRI examination was conducted using ECG-gated sequences. Dynamic images were taken orthogonally to the longitudinal heart axis, from the apex to the base, covering all ventricles and atria for the acquisition of prospective data (Figure 1).

Figure 2 shows 6 multislice-multiphase images between the end-diastolic and end-systolic phases.

The contour of the ventricular epicardium and endocardium and the outline of the left ventricular papillary muscles were defined during the postprocessing of the MRI images (Figure 3).

The progressive changes of volumetric and wall thickness data of the heart cycle are depicted in Figure 4.

The first phase is synchronic with the ECG R-wave, as it constitutes the beginning of the isometric contraction. Ventricular volume is maximum at this point. As the ventricular diastole ends (phase 5–6), the phase of isometric relaxation begins. From a methodological point of view, it is not necessary to follow the entire heart cycle in order to characterize the heart function. It is sufficient to cover the phase from the end-diastole to the end-systole.

Characteristic data concerning lamb heart performance are shown in Table 1, which demonstrates the mean values determined during the heart cycle.

CT scanning was performed on the lambs immediately after the MRI examination and during the same narcosis.

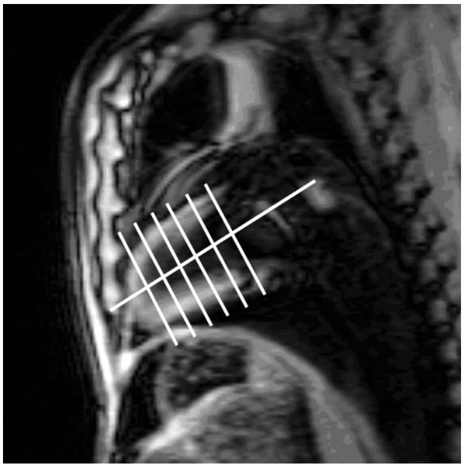

Figure 1. Sagittal localization image with the marking of the longitudinal axis and the levels of dynamic images.

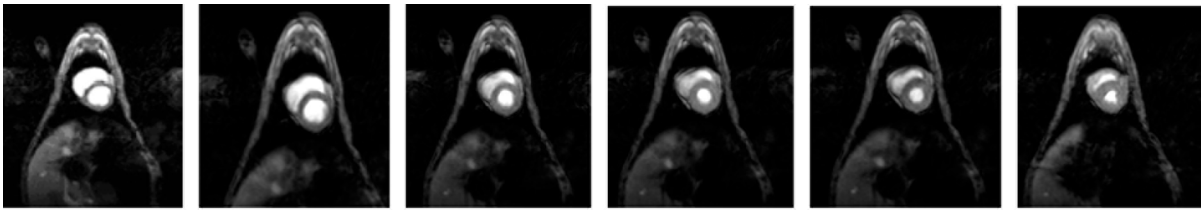

Figure 2. Multislice-multiphase images taken between the end-diastolic and end-systolic phases 9 mm below the level of the atrioventricular valve.

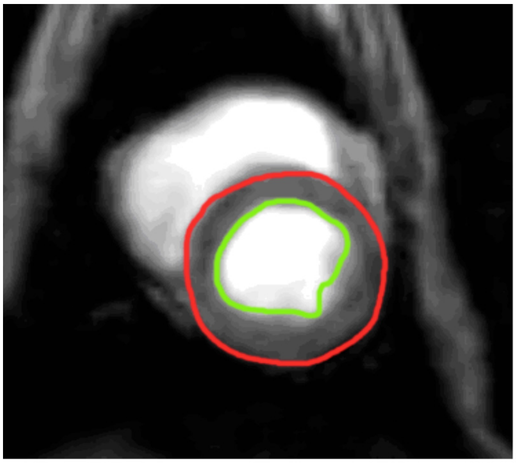

Figure 3. Transversal image with the marking of the left ventricular endo- and epicardium.

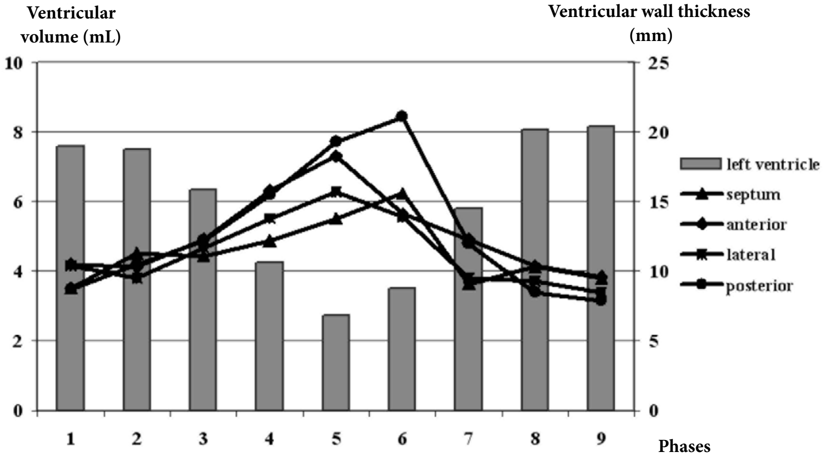

Figure 4. Left and right ventricular volume and the alteration of the left ventricular wall thickness measured at different locations during a heart cycle in a representative animal.

First, total body muscle volume was measured, and then body surface was measured from the consecutive cross-sectional images. CO related to body surface value (dm^2/(L min^{-1}) was estimated following the joint interpretation of MRI and CT data. The so-called relative CO value (expressing the relationship between heart performance and skeletal muscle volume in sedentary conditions (dm^3/(L min^{-1})) was also obtained (Table 2).

4. Discussion

When comparing our results with available data from the literature, the results obtained by ECG should also be considered, and the paucity of such results should be taken into account. However, it must be stressed that no ECG-based cardiac data for sheep in the normal physiological state are available in the literature. The available data concerning the in vivo model have arisen from medical

Table 1. Some characteristic data concerning lamb heart performance.

	LVEDV, mL	LVESV, mL	LVSV, mL	LVEF, %	LV mass, g	HR, beats/min	CO, L/min	Septum, mm	Anterior, mm	Lateral, mm	Posterior, mm
Average	43	11.2	31.8	77.9	63.5	89	2.75	13.6	11.7	11.7	14.2
SD	5.6	1.8	3.8	1.8	5.6	12	0.16	0.0	0.1	0.3	0.1

LVEDV: Left ventricular end-diastolic volume, LVESV: left ventricular end-systolic volume, LVSV: left ventricular stroke volume, LVEF: left ventricular ejection fraction, LV mass: left ventricular mass, HR: heart rate, CO: cardiac output.

Table 2. Related cardiac output data.

	Muscle volume, dm^3	Body surface (BS), dm^2	CO related to BS, $dm^2/(L\ min^{-1})$	Relative CO, $dm^3/(L\ min^{-1})$
Average	6.4	50.3	18.3	2.3
SD	0.79	5.5	3.1	0.26

experiments, where surgically operated animals were used. The findings of Qin et al. (20) originated precisely from that kind of experiment. They found that left ventricular end-diastolic volume, left ventricular end-systolic volume, and left ventricular SV values were 65 ± 24, 30 ± 11, and 35 ± 11 mL, respectively, in the case of sheep with an average body weight of 40 kg (20). They concluded that the accuracy of the method can be verified by cardiac MRI as the gold-standard method. Psaltis et al. (21) measured a slightly higher left ventricular end-systolic volume (37.1 ± 8.8 mL) and a CO value of 3.8 ± 0.6 L/min by applying cardiac MRI on Merino sheep (51 ± 8.1 kg). These results are in accordance with our data, taking the live weight difference (20 vs. 40 kg) into account.

The average 73% ejection fraction value is remarkably higher than the corresponding data of the Mangalica pig (57%), the intensive meat type pig (53%) (14), and the giant turkey (51%) (15). Lower ejection fraction refers to unfavorable hemodynamic characteristics of the heart. The contraction values (septal, anterior, lateral, and posterior: 62%, 69%, 54%, and 58%, respectively) calculated from the measured wall thickness data are characteristic of the condition of the myocardium. Cardiomyopathy or local circulatory failures cause a functional anomaly of muscle fibers and decrease contraction values. Deviation from normal ventricular volume and contraction data is of high diagnostic value.

The average heart rate of 89 bpm can be interpreted as a normal physiological condition (22). The estimated CO value, arising from the SV and heart rate, is one of the most important measures of heart performance in human medical practice. In humans in a sedentary state, approximately 16% of cardiac output is needed to supply the skeletal musculature (23). According to Meyns et al. (24), the perfusion of the sheep muscle is 6 ± 3 mL/g of tissue. Considering the measured average of 6.4 dm^3 muscle tissue and the estimated 2.75 L/min CO value, altogether 14% of total CO supplies the musculature of the examined lambs under narcosis. The estimated CO value is in good accordance with the corresponding data (3.4 L/min) reported by Geens et al. (13). Cardiac output redistribution was studied in a basic experiment carried out by Hales et al. (25) on a Merino model. According to the authors, the competition between skin and muscle for blood flow during exercise results in lower skin perfusion, which could be critical in the case of heat stress.

In human medicine, cardiac output value related to body surface is widely used to characterize metabolic rate. In our study, the respective rate was 18.4 $dm^2/(L\ min^{-1})$, which is considerably lower than the corresponding value of the BUT Big 6 turkey at the same live weight of 20 kg (15).

The developed "relative cardiac output value" expresses the relationship between lamb heart performance and skeletal muscle volume in sedentary conditions. In general, the physical load and/or stress-induced movements of animals lead to significant changes in their blood flow distribution. An example of this phenomenon was studied in ewes by Animut and Chandler (26), who described a 25% decrease in the mammary blood flow during treadmill exercise. In contrast, Segers et al. (17) developed a purely mathematical model for the estimation of SV during strongly differing hemodynamic conditions in sheep. At maximum O_2 consumption, 87% of cardiac output supplies the skeletal muscles in miniature pigs (27). The measured average relative cardiac output value (2.3

dm^3/(L min^{-1}) is similar to the corresponding rate in the native slow-growing Mangalica pig (2.8 dm^3/(L min^{-1}) at 30 kg live weight) (14). This favorably low value signifies a well-balanced cardiovascular system that is characteristic of this semiintensive sheep breed.

In conclusion, our methodological experiment developed a dynamic MRI protocol of the sheep heart. It elaborated the preconditioning and the specific details of ECG measurement and MRI. The above method, combined with CT imaging process of the entire body, gives a unique opportunity to study the skeletal musculature and heart performance in a quantitative and noninvasive manner.

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