EFFECTS OF TRANSPORT ON THE METABOLISM OF HORSES

Efectos del transporte en el metabolismo de equinos

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ABSTRACT

This study assessed the effect of the duration of the transport period on the acid-base equilibrium, energy profile and blood gases of 146 creole equines (CE), 66 of them females and 80 males. Animals, were divided into three treatment groups according to three different transport periods: 52 CEs were not transported and their data were considered as the baseline level (GB); while 49 were transported for two hours (h) (T₂), and the other 45 for 11 h (T₁₁). A marked hemodynamic profile decrease (P < 0.05) in blood pH from group T_{11} (7.23 ± 0.01) and gave rise to a condition of acidemia. Also, a significant increase (P<0.05) was observed in the concentrations of pCO₂, glucose and lactate, as well as in the percentage of hematocrit in group T₂ compared to group T₁₁. The conclusion reached on the basis of these results was that transport periods greater than 2 h cause hyperglycemia, hypercalcemia, hyperlactatemia, hyperpotasemia, metabolic acidosis and a more severe degree of dehydration in creole equines.

Key words: Stress, transport, equines, hemodynamic profile.

RESUMEN

El presente estudio evaluó el efecto de la duración del transporte sobre el equilibrio ácido-base, perfil energético y gasometría sanguínea de 146 equinos criollos (EC): 66 hembras y 80 machos. Los animales fueron distribuidos en tres tratamientos de acuerdo al periodo de transporte: 52 EC no fueron transportados y sus mediciones fueron consideradas como niveles basales (GB), 49 EC fueron transportados durante 2 horas (h) (T_2) y 45 EC fueron transportados durante 11 h (T_{11}) .

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Se observó una disminución significativa (P < 0,05) en el pH sanguíneo, siendo esto más evidente en el grupo T_{11} (7,23 ± 0,01) originando un estado de acidemia. Asimismo, se observó un incremento significativo (P < 0,05) en las concentraciones de pCO₂, glucosa, lactato y porcentaje de hematocrito en el grupo T_2 comparado con el grupo T_{11} . En base a los resultados obtenidos se concluye que, periodos de transporte mayores a 2 h originan hiperglucemia, hipercalcemia, hiperpotasemia, acidosis metabólica y un mayor grado de deshidratación en equinos criollos.

Palabras clave: Estrés, transporte, equinos, perfil hemodinámico.

INTRODUCTION

Currently, many researchers study stress in animals under different approaches [4, 9, 12, 15, 18, 19, 27, 28]. Transport is considered one of the principal stressors in animals, and it is associated with negative effects on their health and meat yield being consequently considered as an element that significantly alters animal welfare [2, 3, 14, 16]. At present, most of the horse (Equus ferus caballus) meat consumed in Mexico comes from injured, sick or old animals, mostly from different towns, circuses or auction houses [1, 8, 27, 28]. The methods of transport and handling prior to slaughter are the main causes of injury, stress and bruising in equines whose meat is destined for human consumption [11]. Generally speaking, transport is a stress factor in horses that can lead to such conditions as the reactivation of salmonellosis [17] increased heart rates, presence of ascorbic acid in the plasma, and increased concentrations of cortisol [5, 13, 23]. Given the large number of factors that can alter the homeostasis of horses during transport, this study assessed the effect of two different duration periods of transport on the acid-base balance, energy profiles,

and blood gas exchanges, with the purpose of establishing an integral physiometabolic profile of the equines that arrive at the abattoir.

MATERIALS AND METHODS

Experimental handling

This study was conducted at a slaughterhouse located in Central Mexico. To assess alterations on the acid-balance equilibrium, energy profile and blood gases of creole equines (CE) an observational, transversal study was designed. This included 146 CE (66 females, 80 males) destined for slaughter, whose meat was to be used for consumption human. On the day prior to transport, blood samples were obtained from a puncture wound in the jugular vein of 52 randomly chosen horses, with 3 mL syringes and 21Gx 32 mm needles. Samples were collected in less than 10 s using syringes previously impregnated with lithium heparin (1000 UI) in order to impede any possible modification of blood values. Sampled animals were at rest and had access to food and water. These results were considered as the baseline treatment (BL). Another sample of horses was randomly assigned to other two treatment groups: two hours (h) transport duration (T2), and 11 h transport duration (T₁₁), over distances of 95.00 and 641.98 km, respectively. Shipping in both groups was performed using gentle handling techniques and setting embarkment ramps at an inclination of 45°. Transport vehicles had no bedding, and the space available for each animal was 0.80 m² for both groups. No food or water was supplied during the entire trip. It is important to remark that the transport conditions assessed in this study are those routinely practiced in Mexico for culled equines. The average values for relative humidity and ambient temperature during the trip in this region of central Mexico range from 35-38% and 18-24°C, respectively [26].

The different treatment regimes were distributed randomly according to transport period, as shown in TABLE I.

Energy profile, acid-base imbalance and blood gases

Upon arrival at the slaughterhouse, the equines were unloaded using similar ramps than at embarkment and herded into the lairage pens. Handling was gentle and no sticks or electrical prods were utilized to move the animals. Prior to entering the lairage pen, the horses were led through a handling area where their weight was recorded on a livestock scale (Reyca Industrial, S.A. de C.V., Básculas & Balanzas, México). At that moment blood samples were obtained following the method described for BL horses. All blood samples were collected by trained veterinary surgeons who strictly followed the guidelines for the ethical use of animals [21] and the procedures for the use and care of animals outlined in the Mexican Regulations for the use of experimental animals [20].

Otic temperatures were measured with an ear thermometer (Braun ThermoScan® IRT 4520, Germany) immedi-

TABLE I
DISTRIBUTION OF TREATMENTS ACCORDING
TO TRANSPORT PERIOD

Treatment	Number of Creole Equines and Transport Period
1 (G _B)	52 Equines (baseline levels)
2 (T ₂)	49 Equines (2 h)
3 (T ₁₁)	45 Equines (11 h)

ately after obtaining blood. Each blood sample was analyzed (GEM Premier 3000, Instrumentation, Laboratory Diagnostics, United States/Italy) to evaluate the following critical blood variables: hematocrit (%), glucose (mg/dL), electrolytes [Na⁺, K⁺ and Ca⁺⁺ (mmol/L)], levels of lactate (mg/dL), partial pressure of carbon dioxide [pCO₂ (mmHg)] and oxygen [pO₂ (mmHg)].

Statistical analysis

To analyze the data, the study used an analysis of variance (ANOVA) for a linear model (PROC GLM; SAS, 2004) which considered the blood variables as dependent variables, and treatments (BL, T_2 and $T_{11})$ as independent variables. When numerical differences were detected, a multiple comparison Tukey test was utilized (P=0.05) to compare the means among treatments. To assess the correlation between blood parameters, Pearson correlation coefficients were calculated (P=0.05). Lactate, glucose, hematocrite, pH, electrolytes, and blood gases were summarized as mean \pm SD. The investigators who did the evaluation and collected the study outcomes were not aware of the treatments and did not participate in the selection of animals or in the data analysis. The investigator who did the analyses was not aware of the treatments.

RESULTS AND DISCUSSION

TABLE II shows the means and standard error for the metabolic profiles, the acid-base balance and blood gas analyses for the equines at baseline values and after transport during two and eleven h.

A state of hypercapnia was observed [pCO₂ (46,36 \pm 1,37 mmHg)] in the equines that were transported during 2 h (T₂), as they showed a significant pCO₂ increase (P < 0.05) in comparison to horses that were transported for 11 h (T₁₁). However, the group of equines that underwent a longer transport period (T₁₁) showed a state of acidemia (pH 7,23) that was lower (P < 0.05) when compared to pH values shown by horses that experienced the short transport period (T₂). With regards to energy metabolism, a condition of hyperglycemia was detected (166,16 \pm 5,64 and 140,55 \pm 4,13 mg/dL) in both groups, as well as a state of hyperlactatemia (41,18 \pm 2,80 and 24,51 \pm 2,16 mg/dL) that showed lower values (P < 0.05) in the group of equines T₂.

TABLE III shows correlation values (P < 0.001) for blood variables in T_2 , equines. Values show that pCO₂, lactate and glucose levels are negatively correlated to pO₂, pH and pO₂ respectively. Associated findings included positive correlations between pCO₂ and glucoselevels and between lactate and Na⁺concentrations.

TABLE IV shows correlation coefficients (P < 0.001) for blood variables in T_{11} equines. Values show, that lactate and glucose levels presented negative correlations with pO₂ and K⁺. In addition, positive correlations coefficients between lactate and glucose levels, and between glucose and concentrations of Na⁺, were observed.

The results of this study indicate that duration of transport time has an effect in CE metabolic balance, acid-base balance, mineral balance, and degree of dehydration. It is important to point out that a transport period of just 2 h is sufficient to trigger hemodynamic changes and to produce hyperglycemia, hypercalcemia, hyperlactatemia, hyperpotasemia, a significant

decrease in blood pH, and an increase in the degree of dehydration.

The hemodynamic alterations observed in the equines exposed to different transport periods can be explained by the stress that the animals experience as a consequence of such factors as hunger, fatigue, ambient conditions, mixing with strange animals and rough handling, that altogether cause a metabolic disequilibrium [10] due to an increase in their basal metabolism leading to greater cardiac expenditure, higher oxygen consumption and higher body temperatures; as well as reduced pH and accumulation of lactic acid.

The significant increase (P < 0.05) in pCO $_2$ concentrations in the equines transported for a short period (T $_2$) caused a state of hypercapnia. In certain situations, such as transport, pCO $_2$ in the alveoli and corporal liquids both rise. When the CO $_2$ bonds with the water, through a process called carbonic anhydrase, it is transformed into carbonic acid, a weak acid that partially dissociates into bicarbonate [6]. With respect to

TABLE ||
MEANS AND STANDARD ERROR FOR OTIC TEMPERATURE AND BLOOD IN EQUINES TRANSPORTED
DURING FOR 0 (GB), 2 (T₂) AND 11 (T₁₁) HOURS

Variables _	Treatments		
	G _B (n=52) Med ± ES	T ₂ (n=49) Med ± ES	T ₁₁ (n=45) Med ± ES
рН	7.47 ± 0.00^{a}	7.37 ± 0.01^{b}	$7.23 \pm 0.00^{\circ}$
pCO ₂ (mmHg)	41.15 ± 0.36°	46.36 ± 1.37 ^a	43.53 ± 0.89 ^b
pO ₂ (mmHg)	39.40 ± 0.31^a	25.57 ± 1.74 ^b	28.82 ± 1.80 ^b
Na⁺ (mmol/L)	132.67 ± 0.65 ^b	140.51 ± 1.20 ^a	139.55 ± 0.78 ^a
K⁺ (mmol/L)	3.20 ± 0.03^{b}	3.32 ± 0.06^{a}	3.33 ± 0.07^{a}
Ca ⁺⁺ (mmol/L)	1.29 ± 0.00^{b}	1.31 ± 0.01 ^a	1.34 ± 0.02^{a}
Glucose (mg/dL)	$82.48 \pm 0.55^{\circ}$	166.16 ± 5.64 ^a	140.55 ± 4.13 ^b
Lactate (mg/dL)	11.25 ± 0.51°	41.18 ± 2.80 ^a	24.51 ± 2.16 ^b
Hematocrit (%)	$38.17 \pm 0.46^{\circ}$	43.06 ± 0.86 ^a	40.60 ± 0.97^{b}

 $^{^{}a, b, c}$ Different literals in the same row indicate significant differences, Tukey (P < 0.05). n: number of equines sampled. SE: standard error. Treatments: GB= group with baseline levels, the equines were at rest with water and food. To any T_{11} groups with 2 and 11 h of transport time, respectively.

TABLE III
PEARSON CORRELATION COEFFICIENTS OF BLOOD VARIABLES IN 49 EQUINES TRANSPORTED DURING 2 h

Dependent variable (y)	Independent variable (x)	R	P Value
DCO (***** U**)	PO ₂ (mm Hg)	-0.593	<.0001
PCO ₂ (mm Hg)	Glucose (mg/dL)	0.543	<.0001
Lactate (mg/dL)	рН	-0.582	<.0001
	Glucose (mg/dL)	0.827	<.0001
	Hematocrit (%)	(3)	<.0001
01	PO ₂ (mm Hg)	-0.718	<.0001
Glucose	Na ⁺ (nmol/L)	0.551	<.0001

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Dependent variable (y)	Independet variable (x)	R	P Value	
	PO ₂ (mm Hg)	-0.587	<.0001	
Lactate (mg/dL)	Glucose (mg/dL)	0.567	<.0001	
	K ⁺ (mmol/L)	-0.416	0.0044	
	PO ₂ (mm Hg)	-0.555	<.0001	
Glucose (mg/dL)	Na⁺ (mmol/L)	0.420	0.0041	

TABLE IV
PEARSON CORRELATION COEFFICIENTS OF BLOOD VARIABLES IN 45 EQUINES TRANSPORTED DURING 11 h

the increases observed in the concentrations of Na^+ and the percentage of hematocrit, Dibartola [7] mentions that hypercapnia has metabolic consequences that include the retention of sodium and water, possibly as a result of an increased liberation of the antidiuretic hormone, an increase in the secretion of cortisol, and the activation of the renin-angiotensin system. In relation to the state of hyperglycemia observed in equines transported for a short period (T_2) , Cunningham y Bradley [6] affirm that the organism under severe stress will secrete large quantities of catecholamines, adrenaline and norepinephrine, which regulate metabolism during stress.

K⁺ (mmol/L)

Duration of transport also contributes to increases in lactate concentrations. Stull and Rodiek [23] found an increase in lactate concentrations in equines between three and six h after transport -i.e., following unloading- suggesting that anaerobic metabolism took place, which is an indicator of minimal muscular fatigue due to transport. In addition, Stull [22] reports that lactate, measured as an indicator of fatigue, showed elevations in horses transported for long (20-27 h) and medium periods (16-23 h), compared to short periods (less than 6 h). During this study, an increase in lactate concentrations was also observed, which caused hyperlactemia in both groups. The observed increase was even larger in equines transported for just 2 h (T₂), compared to horses that were transported for 11 h (T₁₁), a finding that coincides with the results of Werner and Gallo [24, 25], whom observed that equines transported for 59 minutes showed increases in blood lactate concentrations that were attributed to muscular fatigue. Due to the fact that there are few studies of equines in Latin American countries that have assessed the degree of stress that these animals experience during transport to the slaughterhouse, the results of this research on the effects of transport will help to better understand the cause-effect relationships involved in this process, and will serve as a basis for improving the welfare of equines during transport in Mexico.

CONCLUSION

The transport periods (2 and 11 h) evaluated in this study caused hyperglycemia, hypercalcemia, hyperlactatemia, hyperpotasemia, acidosis and increases in the percentage of

hematocrit. Thus, in equines, transport represents a stress factor that causes alterations in the animals' energy metabolism, gas exchanges, blood minerals, and acid-base balance.

<.0001

-0.636

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