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Effects of altitudinal floor on nutrient digestibility, animal´s performance, visceral organ parameters and energy efficiency on guinea pigs.

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EFFECTS OF ALTITUDINAL FLOOR ON NUTRIENT DIGESTIBILITY, ANIMAL´S PERFORMANCE, VISCERAL ORGAN PARAMETERS AND ENERGY EFFICIENCY ON GUINEA PIGS

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RESUMEN

Se realizó una investigación para evaluar el efecto del piso altitudinal en la digestibilidad de nutrientes, eficiencia energética, desempeño productivo y masa de órganos viscerales en cobayos. Veinte cobayos machos con un peso inicial de 1. 011 ± 0. 096 kg fueron seleccionados para un diseño experimental cruzado. Los animales fueron mantenidos en corrales (2 cobayos por corral) durante la fase de digestibilidad que tuvo 2 períodos con una duración total de 25 días. Existieron 2 tratamientos durante el experimento, el piso altitudinal de 2986 m.s.n.m. (metros sobre el nivel del mar) y el piso altitudinal de 2480 m.s.n.m. y existieron 5 corrales en cada sitio. Para el período 1 de digestibilidad, los animales fueron asignados al azar a uno de los dos tratamientos. Una vez finalizado el período 1 de digestibilidad, los animales que se encontraban a 2986 m.s.n.m. fueron cambiados al sitio mantenido a 2480 m.s.n.m. Durante toda la fase de digestibilidad, los cobayos fueron alimentados con 45 gramos de alfalfa para cubrir con los requerimientos energéticos para mantenimiento. Al final de la fase de digestibilidad, un animal de cada corral fue sacrificado para determinar la masa de ciertos órganos viscerales, así como su peso final. Una vez finalizada esta fase, inició la fase de desempeño productivo en donde se realizó un diseño completamente al azar y los cobayos fueron mantenidos en el mismo piso altitudinal al que permanecieron en el período 2 de digestibilidad. Se alimentó a los cobayos con alfalfa y agua ad libitum. Al final de la fase de desempeño productivo, el resto de animales fueron sacrificados y la masa de órganos viscerales fue registrada. El consumo energético y el consumo de materia seca fue mayor en animales mantenidos a 2986 m.s.n.m. en comparación a los animales mantenidos a 2480 m.s.n.m. (P<0. 001). La energía metabolizable tuvo una tendencia a ser menor en los animales mantenidos a mayor altura (P=0. 053). La digestibilidad de nutrientes fue menor en los cobayos mantenidos a 2986 m.s.n.m. en relación a los cobayos mantenidos a 2480 m.s.n.m. La masa del hígado, riñones y bazo fue mayor en los animales mantenidos a 2986 m.s.n.m. vs. 2480 m.s.n.m. (P<0. 012). La masa del corazón tuvo una tendencia a ser mayor en los animales mantenidos a 2480 m.s.n.m. (P=0,060). El peso final, ganancia diaria de peso y conversión alimenticia fue menor a mayor altura (P<0. 002). Los resultados encontrados sugieren que se requiere elevar un 7% adicional de energía metabolizable en cuanto a los requerimientos energéticos para animales mantenidos a un piso altitudinal mayor que en este experimento fueron 506 metros.

Palabras clave: Altitud, cobayo, digestibilidad de nutrientes, masa de órganos viscerales

ABSTRACT

An experiment was conducted to evaluate the effect of altitude on nutrient digestibility, energy efficiency, performance, and, visceral organ mass by guinea pigs. Twenty male guinea pigs (initial BW 1. 011±0. 096 kg) were selected in a crossover design experiment, maintained at metabolic cages (2 animals per cage) during a total digestibility period of 25 d (2 periods of 13-d). Animals were randomly assigned at 1 of 2 altitudinal sites, 2986 and 2480 m. above the sea level (m.a.s.l.; 5 cages per altitude). Animals were fed 45 g of alfalfa (DM) to meet energy requirements at maintenance levels. At the end of the digestion phase, an animal from each cage was slaughter to determine visceral organ mass. A subsequent performance phase was evaluated as completely randomized design, and animals were kept at the same altitudinal floor in which they ended period 2 of the crossover period. Animals were fed *ad libitum* with alfalfa. At the end of the performance phase, all remaining animals were slaughtered and visceral organ mass was measured. Energy intake and Dry matter, were increased by animals at 2986 compared to animals at 2480 m.a.s.l. (P<0. 001). Metabolizable energy tended to be lower for animals kept at 2986 m.a.s.l. (P=0. 053). Nutrient digestibility was lower for animals kept at 2986 compared to 2480 m.a.s.l. Liver, kidneys and spleen mass were greater for animals maintained at 2986 m.a.s.l. (P<0. 012). Heart mass tended to be greater for animals kept at 2480 m.a.s.l. (P=0,060). Final BW, ADG, and feed conversion rate was decreased by animals fed at 2986 m.a.s.l. (P<0. 002). Results from this experiment suggest a novel approach to determine Energy efficiency as affected by altitudinal site. Data from this experiment evidenced a 7% increase on energy requirements on ME for animals kept 506 m.a.s.l. higher. Further research is required to apply to other biological models.

Key words: Altitude, guinea pig, nutrient digestibility, visceral organ mass

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INTRODUCTION

The Andean region is a mountainous area on South America, including countries such as Venezuela, Colombia, Ecuador, Peru, Bolivia and Argentina, with altitudinal floors that varies from 2,000 to 4,000 m.a.s.l. Livestock production is based on grazing production, supplemented with crop residues and agricultural by-products. Previous studies on cattle had shown the effects of altitude on animal´s performance. Animals kept under high altitude conditions have shown Brisket disease also known as High Altitude Disease, a noninfectious cardiac pulmonary condition characterized by edema in the brisket and lower body (Williams, 2012). Geographical Regions at 1800 meters above sea level can express pulmonary arterial pressure as an indicator of risk of developing hypoxia related to susceptibility to pulmonary hypertension and Brisket disease (Crawford et al., 2016; Culbertson et al., 2016).

The percentage of oxygen in inspired air is constant at different altitudes but the atmospheric pressure influences partial pressure of inspired oxygen. At higher altitudes, atmospheric pressure drops and decreases partial pressure of inspired oxygen and hence the pressure for gas exchange in the lungs. Therefore, it will result on a cascade of effects on mitochondria, the final destination of the oxygen (Peacock, 1998). The cellular response to a low Oxygen tension (Hypoxia) is mediated by Hypoxia inducible factors, oxygen sensitive transcription factors that allow adaptation to hypoxic environments. Some adaptation mechanisms are hyperventilation, pulmonary vasoconstriction, polycythemia and respiratory alkalosis. Most mammals increase their hematocrit and alveolar ventilation rate when there is a chronic hypoxemia. These helps animals to maintain normal blood oxygen levels in spite of a reduction in arterial oxygen tension (Shay and Simon, 2012; Costanzo, 2014).

In spite of some evidence related to physiological effects on animals kept at highland conditions, limited research about energy requirements and nutrient utilization on high altitudes is available. Therefore, the objective of this study was to evaluate the effects of altitudinal floor on nutrient digestibility, animal´s performance, visceral organ mass and energy efficiency by guinea pigs.

MATERIALS AND METHODS

All procedures involving live animals were approved by the Universidad San Francisco de Quito Bioethics Committee (protocol number CEUAID-USFQ001).

Animal Management and Treatments

Twenty male Peruvian line guinea pigs (*Cavia porcellus*) were used on this experiment. Animals had an initial BW of 1.01 kg ± 0.10, and were 4 weeks old. Animals were placed (2 per pen) on metallic and wood pens (0.60 m. long x 0.85 m. depth x 0.40 m. height).

Animals were randomly assigned to one of two altitudinal sites (2480 or 2986 m. a. s. l.; 5 block per site). The experiment had 2 digestibility periods. At the end of period 1, animals kept at 2986 m.a.s.l. went to 2480 m.a.s.l. and vice versa. After the digestibility phase, there was a performance phase where animals were kept on the altitudinal floor they ended the digestibility phase.

Experiment Management

Digestibility Period:

During the digestibility period, animals had an adaptation period of at least 7 days. And a collection period of 5 days. During the adaptation and digestibility periods, animals were fed 45.34 g of alfalfa (DM basis) and water *ad libitum*. Energy was supply to meet maintenance requirements of the animals following this Eq.:

$Energy\ maintenance\ (Kcal/d) = 136\ \times\ live\ weight\ (kg)^{0.75}$ (NRC, 1995)

Temperature and relative humidity were recorded (BoecoGermany, Boe 330, Hamburg ± 1 mg) daily at 6 different times through the day.

Animal body weight was recorded before and after the digestibility period (Camry Ek3550, City Industry, CA, scale readability ± 1 g).

Digestibility Measurements

Feces excreted were collected quantitatively for 5 consecutive days. Total excreted feces were weighted, thoroughly mixed and an aliquot of 100 g was stored at -20° C until composite. Composite fecal samples were dried at 60 ° C for 72h. Samples were grounded in a mill (Mark Thomas Scientific, 3383 -LI0, Swedesboro, NJ) to pass a 1mm screen and stored for laboratory analysis.

Estimates of the approximate quantity of unconsumed feed remaining were recorded daily for each pen. Fecal and feed samples were assayed for ash (Method 942.05; AOAC, 1995), CP (Method 988.05; AOAC, 1995), fat (Method 920.39. AOAC, 1995), crude fiber (Method 962.09. AOAC, 1995) and energy (Calorimetric bomb).

Urine was collected daily for 5 consecutive days into plastic containers containing 100 mL of 3 *M* HCl to keep urine pH ≤ 2. Collected urine was thoroughly mixed, volume and weight were recorded. Total urine output was refrigerated until composited and stored at -80 ° C until analysis. Energy was determined using a calorimetric bomb.

Performance period:

For performance period, animals were assigned to a completely random design. After the digestibility period, animals kept on highland conditions remained at 2986 m.a.s.l. Meanwhile, animals who ended at 2480 m.a.s.l. were kept on the same altitudinal floor. During this period, on both sites, animals were fed with alfalfa and water *ad libitum* for 19 d. Dry matter intake, ADG and feed conversion ratio were recorded.

Slaughter procedure

Three animals from different pens from each altitudinal floor were randomly selected for slaughter procedure at the beginning of the performance period. At the end of this period, remaining animals were slaughtered. They were stunned with a captive bold, following exsanguination and evisceration. Visceral organ mass was registered. Carcass and organ´s mass including heart, lungs, stomach, small and large intestine, liver and spleen were recorded with an analytical balance (Mettler Toledo, p6203-S, Columbus, IO ± 1 mg).

Organ Volume

Volume of the heart, stomach, lungs, kidneys, spleen, small and large intestine, cecum and liver were measured using the water displacement method. Each organ was submerged in a graduated cylinder containing a prefixed amount of distilled water. A picture was taken of the cylinder before and after dropping the organ in it. Water displacement was accurately measured in the images using ImageJ Software.

Statistical Analysis

Data from the experiment were analyzed using the MIXED procedure (SAS Institute, Inc., Cary, NC). Sources of variation included animal treatment and treatment sequence. Significant difference was declared at $P \leq 0.05$.

RESULTS AND DISCUSSIONS

During the experiment, the average temperature at 2986 m.a.s.l. was 16°C and at 2480 m.a.s.l. was 15.35°C. The average relative humidity was 74.24% at 2986 m.a.s.l. and 75.15% at 2480 m.a.s.l.

Digestibility phase

Performance: On the digestibility phase, initial full BW was not different across altitude site (P=0.349). Final body weight was significant different between treatments (P=0.015). Animals at 2480 m.a.s.l. had a heavier final BW in comparison to animals at 2986 m.a.s.l (Table 1). Dry matter intake was significantly different across treatments (P<0.001). Animals at 2986 m. a. s. l. had a higher DMI.

Energy partitioning: Energy intake was significantly different across treatments (P=0.002), animals at 2986 m. a. s. l. had a higher energy intake (Table 1). Fecal energy was not significantly different between animals kept at 2986 and 2480 m.a.s.l. (P= 0.259). No significantly differences were noted among treatments for digestible energy (P=0.128). Nevertheless, animals kept at 2480 m. a. s. l. had higher urinary energy (P<0.001). Metabolizable energy was not altered by treatments, (P=0.053). Animals at 2986 m. a. s. l. consumed more energy. Related to higher energy requirements in comparison to animals kept on 2480 m. a. s. l. Previously, it has been suggested that hypoxia correlated with high altitudes causes a reduction in feed intake independently of climatic influences and diet (Leiber et al., 2006). The principal mechanism of ATP production in cells is through oxygen chemical reduction during oxidative phosphorylation (Halligan, 2016). Ambient air contains 21% oxygen and the majority of healthy tissues have access to 2-9% and hypoxia is defined as less than 2% (Shay and Simon, 2012).

An altitudinal floor difference of 506 m. will result on a lower availability of oxygen due to a lower barometric pressure. Therefore, at a higher altitudinal floor, animals will have a lower availability of oxygen for gastrointestinal metabolic activities. Humans and animals have developed an oxygen-sensing mechanism that enables cells to adapt and maintain homeostasis under hypoxic conditions. The activation of genes that mediated metabolic adaptation, angiogenesis, energy conservation and erythropoiesis are some of the adaptive mechanisms to hypoxia. As a result of an inadequate oxygen supply, cells shift from an aerobic to an anaerobic metabolism, therefore, glycolysis becomes the primary source of energy production. However, glycolysis produces only 2 mol of adenosine triphosphate (ATP)/ mol of glucose as compared to 36 mol ATP/ mol glucose during aerobic respiration (Lakshman and Bonventre, 2009). Changes in metabolism and methods of ATP production is not the only way in which cells adjust to low oxygen tension. Cells also adaptively alter mechanisms by which biosynthesis of macromolecules occurs, using metabolic intermediates to fuel nucleotide and lipid synthesis (Shay and Simon, 2012).

Additionally, the gastrointestinal tract is very active metabolically (Webster, 1980). Estimates on oxygen consumption have been made in other species such as rats (12%), pigs (24.8), sheep (18.5%) and cattle (17.7%) (Foster and Frydman,1978; Thompson et al., 1978; Huntington and Tyrrell, 1985, Yen et al., 1989). The major biochemical events that consume oxygen in the gastrointestinal tract is for cellular homeostasis and growth, specifically Na+, K+-ATPase activity, protein synthesis and protein degradation (McBride and Kelly, 1990). Suggesting that at higher altitudes, there is less oxygen available for tissues and for gastrointestinal metabolic activities, related to high energy demand for metabolism, as suggested in our study.

Other studies in mice kept on different sites from 1,200 to 3,800 m. a. s. l. revealed that animals living at high altitudes have higher levels of energy demand (Hammond et al.,1999). In other studies, where milk cows have been evaluated under two altitudinal conditions, milk yields at high altitudes were lower than milk yields at low altitudes in Chinese Holstein cows. This can be correlated with nutrient requirements. Chinese Holstein cows at higher altitudes need more nutrients to meet maintenance requirements, especially with regard to energy (Qiao et al., 2009). Published studies in dairy cows at alpine altitudes compared to lowland altitudes revealed increases in energy requirements for maintenance, 0.72 times greater at 2000 m. a. s. l. vs. 400 m. a. s. l. (Berry et al., 2001). Yet, in other published studies, there were opposite results found in sheep (Blaxter, 1978), goats (Forster et al., 1981), and yaks (Han et al., 2003). Energy requirements in these species were not affected by the altitude ranging from 2270 to 4260 m. a. s. l. This can be associated with adaptation of these species to Tibetan plateau (Qiao et al., 2009).

Data from other studies demonstrated that energy required for maintenance in small to medium sized species is usually elevated when animals are exposed to high altitude conditions for 3 to 4 weeks. (Butterfield et al., 1992). Yet, in this study, energy requirements were affected by a difference of 500 m. a. s. l. in a period of less than 3 weeks.

During the digestibility phase, animals kept on 2480 m. a. s. l. were able to gain weight in comparison to animals at 2986 m. a. s. l. This is linked with ME. Metabolizable energy was higher in animals at 2986 m. a. s. l. than animals at 2480 m. a. s. l. even though it was no significantly different, as seen in Table 1. This suggests that animals at a higher altitude required more energy to keep their metabolic demand. As well, at 2986 m. a. s. l., an increase in ME can be associated with a higher heat increase. Suggesting that animals kept at 2986 require 7% more of ME. Presumably, animals at 2480 m. a. s. l. consumed less energy to meet their metabolic demand in comparison to animals at 2986 m. a. s. l. Meaning that at a lower altitude, animals are able to consume energy not only for maintenance but for production too. For further studies, it is recommended to measure heat increment so it can be estimate how much energy did the animal use for maintenance and production.

Urinary energy was higher in animals kept on 2480 m. a. s. l. in comparison to animals kept on 2986 m. a. s. l. (Table 1). One of the reasons why animals at 2480 m. a. s. l. might registered a higher urinary energy is due to a higher water consumption, therefore, there was a higher urine output in these animals. In the present study, water consumption was not registered, however, temperature and humidity was controlled. Thus, no climatic conditions can be attributed with a higher water consumption.

Nutrient intake and digestibility: Table 1 indicates organic matter, crude fiber, crude protein, ether extract and nitrogen digestibilities. Animals fed at 2480 m. a. s. l. registered a higher organic matter, fiber, crude protein and NFE digestibility in comparison to animals placed at 2986 m. a. s. l. Previously, Qiao et al., 2009 did not find any effect of altitude (1600 vs. 3600 m. a. s. l.) on nutrient intake and digestibility on dairy cows.

Performance phase

Performance: As shown in Table 2, initial BW did not have significant differences across treatments. Feed intake did not register significant differences between treatments. At 2480 m.a.s.l. animals had a greater ADG and lower feed conversion ratio (P<0.002; Table 2). Animals at 2480 m. a. s. l. had a greater final BW (P=0.0014), and a greater empty BW compared to animals at 2986 m. a. s. l. (P<0.001; Table 2). Even though, both groups of animals recorded a similar feed intake, animals at a lower altitude were able to gain more weight. Presumably, guinea pigs kept on 2480 m. a. s. l. were able to deposit more nutrients in tissues comparing to animals kept at 2986 m. a. s. l. Animals fed at 2986 m. had greater energy requirements to meet their metabolic demand. This agreed with a published study in cows kept on highland conditions that revealed a lower average daily gain when compared to cows kept on lowland conditions (Donald et al., 1962).

Results from other published studies have reported that the effects of high altitude on animals can be associated with climatic conditions such as ambient temperature and humidity, solar radiation and topographic challenges under grazing conditions (Lieber et al., 2004). Published studies have revealed that cows grazing at high altitudes (2000 m.a.s.l.) showed higher energy requirements in comparison to cows remaining on lowland pastures (400 m.a.s.l.). Cows at high altitudes were estimated to require twice as much energy to meet maintenance requirements compared to lowland conditions. This increase on energy requirements is associated to cold stress. It seems that grazing helps the cows to adapt to the high altitude conditions at a cost of a considerably higher energy expenditure which is compensated with higher feed intake (Christen et al., 1996). However, in our study, both experimental groups, were exposed to controlled temperature and humidity maintaining animals at similar conditions. Therefore, dry matter intake can not be associated with other factors such as temperature, humidity and solar radiation.

Visceral organ mass, volume and density: As shown on Table 2, at the beginning of performance period, visceral organ mass were not different between treatment groups ($P>$ 0.059), except for cecum which was greater in animals kept at 2480 vs. 2986 m.a.s.l. ($P=$ 0.026). At the end of performance period, animals at 2986 m.a.s.l. had greater liver, kidneys and spleen mass ($P \leq 0.013$). Animals at 2986 m.a.s.l. had a greater liver ($P= 0.001$) and kidneys volume ($P= 0.004$) in comparison to animals at 2480 m.a.s.l. There were no significant differences on the other organs collected for mass and volume. There were no significant differences for organs density, except for spleen´s density ($P= 0.0322$). Animals at 2986 m.a.s.l. had a greater spleen density.

Published studies in mice kept at 1200-3800 m.a.s.l. revealed that animals living at high altitudes have greater cardiopulmonary and digestive organs than mice living at lower altitudes (Hammond et al.,1999). In high altitude mice, phenotypically changes in organ mass allow the organism to maintain an adequate oxygen uptake and delivery. Presumably, digestive organs are larger at high altitude mice due to higher feed intakes resulting on higher thermoregulatory costs and decreased ambient temperatures. Also, heart and lung tissue are greater at high altitude mice apparently as a result of a lower oxygen partial pressure. This is a result of a greater lung function to gain enough oxygen, as well as greater heart muscles contractions to pump more blood to the tissues (Hammond et al., 2001).

In the present study, animals kept on highland revealed a greater mass in kidneys and spleen in comparison to animals kept on 2480 m. a. s. l. In addition, kidney volume was higher in animals at a higher altitude. In other studies, animals exposed to higher altitudes revealed a 16% smaller kidney in comparison to mice at a lower altitude (Hammond et al., 2001). Similarly, a study performed in humans, revealed that people who reside at 2220 m. a. s. l. have a lower kidney size and volume (Musa and Abukonna, 2017). A study performed by Beregi et al. (2001) revealed ultrasonographic renal measurements in 31 guinea pigs, however, there is no information about ultrasonographic renal measurements in guinea pigs exposed to high altitudes.

One possible explanation to a greater mass in guinea pig´s kidneys at a higher altitude can be due to an increase of the erythropoietin production cells. The hormone erythropoietin is the principal growth factor that promotes the viability, proliferation and differentiation of erythroid progenitor cells. Erythropoietin blocks apoptosis in the late erythroid progenitors enhancing proliferation and terminal differentiation of their progenies. Therefore, is essential for red blood cells production. The specific stimulus for erythropoietin expression is a fall of oxygen pressure in tissues. Its production increases under hypoxic conditions specifically in the interstitial cells in the renal cortex and outer medulla of the kidney and in less amounts in the liver, brain, spleen and lung. Erythropoietin production depends on the tissue oxygen pressure but is also activated when the arterial oxygen pressure declines or when the oxygen affinity of the blood increases. An increase in erythropoietin will result in erythrocytosis, which is common in high altitude residents (Youssoufian, 1993; Jelkmann, 2011 and Yoon et al, 2011). An increase in red blood cell production will help deliver oxygen to tissues more efficiently. But, an excessive erythrocytosis can result on an increase blood viscosity which impairs tissue blood flow leading to impaired tissue oxygen delivery and other adverse effects named high altitude polycythemia (Peng, et al. 2011 and Yoon et al, 2011). For future studies, height and volume of renal cortex should be analyzed because this will help to determine if erythropoietin is being produce at higher rates as an adaptation to hypoxia in animals kept on higher altitudes. Additionally, histological analysis could be considered as indicator of morphological changes in cells.

Animals at 2986 m.a.s.l. had greater spleen´s density. The spleen has two functionally and morphologically distinct compartments, the red pulp and the white pulp. The red pulp is responsible for filtering blood, removing foreign material and damaged erythrocytes. As well, platelets, iron and red blood cells can be store there. In rodents, hematopoiesis takes place in the red pulp in fetal and neonatal animals. The white pulp is responsible of containing about 25% of the body´s lymphocytes and initiates immune response to blood-borne antigens (Kuper et al., 2002; Nolte et al., 2002 and Balogh et al., 2004).

A study performed in dogs and guinea pigs where the lungs were evaluated at high altitude shown that guinea pig had smaller spleens (0.3% of BW) and had limited capacity of carrying hematological oxygen. Consequently, guinea pigs rely on structural lung growth for gas exchange compensation at high altitudes. Whereas, a dog´s spleen (3% of BW) sequesters and releases erythrocytes into the circulation, an efficient form of reversible blood doping that augments diffusive oxygen transport avoiding the effects of chronic polycythemia. The canine splenic contribution for oxygen transport in chronic hypoxia mitigates the need for alveolar tissue growth, the one that incurs a higher metabolic cost (Ravikumar et al., 2015). Presumably, animals at a higher altitude have a greater density and mass of the spleen due to sequestration red blood cells resulting of the erythrocytosis cause by hypoxia.

On another hand, studies have shown that the liver accounts for approximately 1.5 to 2.5% of BW and it consumes up to 25% of the total oxygen used by the whole body (Huntington and McBride, 1988). According to McBride and Kelly (1990), energy costs are high for five biochemical processes Na+, K+-ATPase activity, protein synthesis, protein degradation, substrate cycling and urea synthesis. It was shown that maintenance of liver protein synthesis alone could consume up to 4.9% of whole-body ATP use, assuming that the liver accounts for 20% of whole-animal ATP use. A considerable amount of cellular energy is consumed for substrate cycles such as glucose/glucose 6 phosphate, glycogen/glucose I –

phosphate, fructose 6-phosphate triacylglycerol/fatty acid, cholesterol/cholesterol esters, AMF´ /adenosine, acetyl CON acetate, fatty acyl CoNfatty acid and glutamine/glutamate (Newsholme and Stanley, 1987). This finding can be associated with a higher metabolic activity that animals at a 2980 m.a.s.l. had due to a higher feed consumption. Consequently, this group of animals had to metabolized more nutrients so they could be deposit in tissues and red blood cells could be produced.

CONCLUSION

Results from this experiment suggest a novel approach to determine Energy efficiency as affected by altitudinal site. Data from this experiment evidenced a 7% increase on energy requirements on ME for animals kept 506 m.a.s.l. higher. Further research is required to apply to other biological models.

Table 1. Effect of altitudinal floor on energy efficiency and nutrient digestibility on digestibility phase.

	Altitude, m.a.s.l.			*P*-value[1]
Item	2986	2480	SE[2]	
IFBW[3] kg,	1.02	1.00	0.026	0.349
FFBW[4] kg	1.01	1.04	0.029	0.015
DMI, g	45.24	44.44	1.344	<0.001
Energy Intake, cal	2059.69	1956.59	127.30	0.002
Fecal Energy, cal	176.20	213.05	23.03	0.259
Digestible Energy, %	91.33	86.82	2.13	0.128
Urinary Energy, cal	48.81	74.67	4.66	<0.001
Metabolizable energy, %	88.72	82.73	2.14	0.053
Organic Matter, %	80.92	88.37	0.81	<0.0001

Fiber, %	67.69	82.61	1.57	<0.0001
Crude Protein, %	87.20	91.90	0.44	<0.0001
Ether extract, %	87.19	91.37	0.75	0.001
Nitrogen Free Extract, %	84.02	89.50	0.937	0.002

[1]*P*-value: Significance level.

[2]SE: Standard error of treatments means (*n=5*).

[3] IFBW: Initial full body weight at the beginning of the experiment.

[4] FFBW: Final full body weight at the end of the digestion period.

Table 2. Effect of altitudinal floor on animal performance and visceral organ mass.

	Altitude, m.a.s.l.			*P*-value[1]
Item	2986	2480	SE[2]	
Initial BW, g	1046.1	1018.4	42.38	0.0656
Final BW, g	1233	1510	41.056	0.0014
ADG	9.86	25.90	2.60	0.0024
Feed Intake, g/d	39.31	37.64	2.018	0.5759
Feed Conversion Ratio	4.30	1.58	0.413	0.0016
d 19				
Visceral organ mass, g/kg EBW				
Empty BW, g	1093.24	1375.77	41.73	<0.001
Heart	4.36	6.10	0.73	0.060
Lungs	12.17	13.64	3.28	0.693
Liver	44.66	34.07	2.70	0.008
Kidneys	15.90	12.40	0.98	0.012
Spleen	2.49	1.71	0.23	0.013
Stomach	11.05	12.32	1.79	0.595

Cecum	21.23	25.87	3.64	0.279
Large Intestine	23.67	22.50	1.77	0.565
Small Intestine	19	18.12	4.30	0.856
Pancreas	2.87	1.90	0.50	0.114

[1] *P*-value: Significance level.

[2] SE: Standard error of treatments means (*n=5*).

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