UNIVERSIDADE ESTADUAL DE MATO GROSSO DO SUL
UNIDADE UNIVERSITÁRIA DE AQUIDAUANA
PROGRAMA DE PÓS-GRADUAÇÃO EM ZOOTECNIA

EFEITO DO SOMBREAMENTO SOBRE O COMPORTAMENTO NA AMAMENTAÇÃO E ÍNDICE DE KLEIBER EM BEZERROS DE DIFERENTES GRUPOS GENÉTICOS

Acadêmica: Letícia Scarelli Rodrigues da Cunha

Aquidauana-MS
Novembro de 2021

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Orientador: Prof. Dr. Dalton Mendes de Oliveira

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APROVADA em 26 de Novembro de 2021.

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(Orientador)

Dr. Marcos Paulo Gonçalves de Rezende
(via videoconferência)

Dr. Gumercindo Loriano Franco
(via videoconferência)

“Não to mandei eu? Esforça-te, e tem bom ânimo; não temas, nem te espantes; porque o Senhor, teu Deus, é contigo, por onde quer que andares”

Josué 1:9-13

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LISTA DE ABREVIAÇÕES

CAN	- Análise canônica
DMM	- Duração média da mamada
GMD	- Ganho médio diário
GPD	- Ganho de peso diário
IK	- Índice de Kleiber
ITU	- Índice de temperatura e umidade
N	- Número de dias por período
NA	- ½ Nelore x ½ Angus
NE	- Nelore
NM	- Número de atos de mamada por dia
NP	- ½ Nelore x ½ Pantaneiro
PF	- Peso final
PI	- Peso inicial
PV	- Peso vivo
TA	- Temperatura ambiente
TºC	- Temperatura do ar em graus Celsius
TCS	- Temperatura corpórea superficial
UR	- Umidade relativa

RESUMO

O Pantanal brasileiro, produtor famigerado de bovinos de corte, é caracterizado por altos níveis de temperatura e umidade, condições que demandam a seleção de animais adaptados para tal. Neste sentido, torna-se interessante o uso de raças tais como a Pantaneiro, naturalizada da região, devido a sua elevada rusticidade e inferior exigência alimentar, porém em risco de extinção; ou a raça Nelore, já amplamente utilizada por responder bem fisiologicamente às intempéries. Além destas, utiliza-se cada vez mais animais Angus na busca por maiores ganhos em produtividade, entretanto, estes não se adaptam bem às adversidades climáticas, o que torna o cruzamento entre espécies interessante. Assim, objetivou-se determinar o efeito do estresse térmico sobre o comportamento de amamentação e desempenho de bezerros de três diferentes grupos genéticos, em ambientes de pastejo com e sem a presença de sombreamento, através do Índice de Temperatura e Umidade (ITU) e Índice de Kleiber (IK). Foram avaliados 25 animais, a partir dos 30 dias de idade distribuídos em: 9 bezerros Nelore (NE), 8 cruzados ½ Nelore x ½ Angus (NA) e 8 ½ Nelore x ½ Pantaneiro (NP), das 7 às 17 horas, em ambientes providos ou não de sombreamento natural. Foram coletados dados de número de atos de mamar por dia (NM), duração média da mamada (DMM), temperatura corpórea superficial (TCS), ITU, ganho de peso e IK. O ITU apresentou diferença ($P<0,05$) entre os ambientes. Animais NA apresentaram maior TCS (33,00±2.31), diferindo ($P<0,05$) de NE (31,71±1,65) e NP (31,38±1,99), e menor ($P<0,05$) atividade de amamentação, sugerindo redução do conforto térmico, entretanto, seu ganho de peso e IK foram superiores, diferindo ($P<0,05$) dos demais grupos genéticos. Já animais NE e NP não diferiram ($P>0,05$) em ganho de peso e IK. Portanto, os resultados sugerem que os bezerros NA, apesar do maior desconforto térmico, utilizaram de mecanismos de termólise para manterem ganho de peso e eficiência alimentar (IK) maiores que os demais grupos. O NP apresentou maior adaptabilidade térmica, além de ganho de peso e IK semelhantes aos animais NE.

Palavras-chave: Adaptação, Bovinocultura, Desempenho, Eficiência alimentar, Índice de Temperatura e Umidade, Pantanal

ABSTRACT

The Brazilian Pantanal, a well-known producer of beef cattle, is characterized by high levels of temperature and humidity, conditions that require the selection of animals adapted for this. In this sense, it becomes interesting to use breeds such as Pantaneiro, naturalized from the region due to its high rusticity and lower food requirement but at risk of extinction; or the Nellore breed, already widely used for responding well physiologically to the weather. In addition to these, more and more Angus animals are used in the search for greater gains in productivity, however these do not adapt well to the weather making the crossing between species interesting. Thus, the objective was to determine the effect of thermal stress on the breastfeeding behavior and performance of calves from three different genetic groups, in grazing environments with and without the presence of shading, through the Temperature and Humidity Index (THI) and Kleiber Index (KI). Twenty-five animals were evaluated from 30 days of age: 9 Nellore calves (NE), 8 crossed ½ Nellore x ½ Angus (NA) and 8 ½ Nellore x ½ Pantaneiro (NP), from 7 to 17 hours, in environments provided or not of natural shading. Data on the number of feeding acts per day (NFA), mean duration of feeding (MDF), body surface temperature (BST), THI, weight gain and KI were collected. The THI showed a difference (P<0,05) between the environments. NA animals presented higher BST (33,00±2.31), differing (P<0,05) from NE and NP, and lower (P<0,05) breastfeeding activity, suggesting a reduction in thermal comfort, however their weight gain and KI were higher, differing (P<0,05) from the other genetic groups. Therefore, the results suggest that NA calves despite the greater thermal discomfort, used thermolysis mechanisms to maintain weight gain and feed efficiency (KI) higher than the other groups. NP animals showed higher thermal adaptability to the environment, in addition to weight gain and KI similar to NE animals.

Key words: Adaptation, Cattle breeding, Food Efficiency, Pantanal, Performance, Temperature and Humidity Index

CAPÍTULO 1 - CONSIDERAÇÕES GERAIS

1. INTRODUÇÃO

O Pantanal brasileiro, reconhecido por ser a maior área alagada do planeta com 4.385.000 hectares inundáveis (CATELLA et al., 1996), tem como principal atividade econômica a produção de bovinos de corte e, por sua variação em seu regime hidrológico, concentra nas regiões alagáveis o sistema de cria, nomeando-a de “berçário” da pecuária, que abastece toda a cadeia produtiva nacional de carne (ARAUJO et al., 2016; ARAUJO et al., 2018).

Além das características hidro territoriais, o Pantanal situa-se na faixa tropical do planeta, recebendo altos níveis de irradiação solar e, consequentemente, apresentando elevadas temperaturas. No ano de 2020, a média anual de temperatura e umidade desta região foi de 25,6ºC e 68,59%, respectivamente (INMET, 2021). Associadas, essas variáveis estimulam o aumento da temperatura corporal, induzindo ao estresse térmico, reduzindo a ingestão calórica e, consequentemente, o ganho de peso (HERBUT et al., 2018).

Dessa forma, a escolha de raças para produção de carne neste cenário demanda características adaptativas às condições climáticas, por isso, muitas vezes utilizam-se animais da raça Nelore, os quais respondem bem fisiologicamente às intempéries (BARBOSA et al., 2017). Outra opção que atende estes requisitos é a raça Pantaneiro, animal naturalizado do Pantanal devido sua alta rusticidade e menor exigência nutricional, e que hoje, apesar das características singulares, corre risco de extinção (REZENDE et al., 2014; ALBUQUERQUE; IANELLA, 2016).

Acompanhando a evolução da pecuária no Brasil, aliado à preconização ascendente de produtos cárneos de melhor qualidade por parte do mercado interno e externo, os produtores vêm utilizando também cada vez mais os animais taurinos, principalmente os da raça Angus, pela diferença superior em ganho de peso e acabamento (DIAS et al., 2015). Entretanto, este grupo genético apresenta dificuldades de adaptação às condições edafoclimáticas brasileiras, por isso, o cruzamento entre taurinos e zebuínos pode ser interessante até mesmo para o produtor pantaneiro, uma vez que une as características de

desempenho e adaptação de ambas as raças (BATTISTELLI et al., 2013), todavia, estudos desse cruzamento sobre as condições do Pantanal ainda precisam ser melhor explorados.

Nesta perspectiva, intensificaram-se os estudos e abordagens acerca da influência térmica sobre os atributos produtivos de bovinos de corte e, para tal, desenvolveram-se equações que preditem com precisão o nível de estresse térmico imposto aos animais (WANG et al., 2018). É o caso do Índice de Temperatura e Umidade (ITU), um dos mais utilizados neste propósito, que classifica em escala o nível de estresse térmico imposto aos animais através dos indicadores de temperatura e umidade (STORTI et al., 2019).

Paralelamente, no sentido de abordar a eficiência em ganho de peso e, posteriormente, relacioná-la ao estresse térmico, utiliza-se de equações como o Índice de Kleiber (IK), alternativo à conversão alimentar, capaz de predizer a eficiência energética dos animais através da relação direta entre o peso metabólico e ganho médio diário (GMD) dos animais, sem a necessidade de mensuração da ingesta (KLEIBER, 1936).

Logo, objetivou-se avaliar o grau de influência térmica sobre o desempenho de bovinos de corte, através das equações de ITU e IK, preconizando o bem-estar animal e o incremento da produção de bovinos de corte.

2. REVISÃO DE LITERATURA

2.1 Perfil adaptativo e produção de bovinos

O perfil de produção de bovinos no Brasil ocorre majoritariamente em sistemas extensivos, ou seja, animais criados a pasto (DIAS et al., 2017). Levando em consideração as características tropicais do país, aliado ao fato de que este sistema de produção é altamente dependente das condições climáticas (MORALES et al., 2020), faz-se essencial o conhecimento das respostas fisiológicas e, consequentemente, produtivas, dos animais frente a estes estímulos.

Em períodos de temperatura elevada, a reação natural dos bovinos é de manter a homeotermia, ou seja, estabilizar o organismo dispondo de recursos fisiológicos de transferência de calor, alcançando melhor conforto térmico. Na prática, esses animais irão procurar locais sombreados, aumentam a ingestão de água e minimizam o consumo de alimentos, elevam a frequência respiratória bem como a sudorese e a vasodilatação periférica (PEREIRA et al., 2017).

Entretanto, estes mesmos mecanismos que os resguardam de um possível evento de estresse térmico, são também responsáveis pelo decréscimo de ganhos produtivos e econômicos (FURTADO et al., 2012), uma vez que há redução de ingestão calórica e consequente balanço energético negativo e possível perda de peso, podendo ainda acarretar em uma pior resposta do sistema imunológico (TAKAHASHI et al., 2009).

Em face do exposto, a escolha de animais de constituição genética que atendam às características produtivas, das quais sejam minimamente influenciadas por fatores climáticos, torna-se relevante. Em caráter comparativo, animais *Bos taurus indicus* (zebuínos) tendem a regular mais eficientemente a temperatura corpórea em situações de estresse térmico comparativamente a *Bos taurus taurus* (taurinos) como a raça Angus, uma vez que a faixa de conforto térmico dos zebuínos varia entre temperaturas mais elevadas (10 a 27°C), enquanto os taurinos habituam-se a temperaturas mais amenas (0 a 16°C) (KAZAMA et al., 2008).

Justifica-se assim a expressiva representatividade da raça Nelore e seus cruzamentos no rebanho brasileiro, pois se adequam muito bem às condições tropicais do país (DIAN et al., 2020), uma vez que apresentam características morfológicas relevantes para tal, como a pelagem curta e cor clara, refletindo maior radiação, e epiderme pigmentada, protegendo-os da incidência ultravioleta (TAKAHASHI et al., 2009).

Ainda no conceito de adaptabilidade climática, a raça Pantaneiro, apesar de herdar material genético de ancestrais taurinos (ISSA et al., 2006), é considerada genuinamente brasileira e completamente aclimatada ao bioma Pantanal (RUFINO JÚNIOR et al., 2014).

Sabendo das prerrogativas de conforto e adaptabilidade térmica de cada padrão racial e intencionando a maximização da produção cárnea, o cruzamento se torna ferramenta interessante na complementaridade entre raças, alcançando

níveis superiores em desempenho e qualidade de carne, incrementando a criação e comercialização de bezerros (BATTISTELLI et al., 2013).

2.2 Índice de Temperatura e Umidade

Como a termorregulação está intimamente relacionada com variáveis climáticas, são diversas as medidas existentes atualmente para mensurar esses efeitos, como temperatura ambiente, umidade relativa do ar, velocidade do vento e incidência de radiação. Entretanto, estes índices sozinhos não são capazes de aferir com acurácia o conforto, ou estresse, térmico, visto que as características morfológicas (coloração e estrutura do pelo e cor da pele) de cada animal podem interferir significativamente no nível de absorção de radiação (PEREIRA et al., 2017).

Por conseguinte, novos métodos de mensuração foram criados, como o Índice de Temperatura e Umidade (ITU), desenvolvido por Thom (1958), que pode representar fidedignamente o estresse ou conforto térmico de um animal através das medidas de temperatura e umidade conjuntamente (temperaturas de bulbo seco e úmido) (STORTI et al., 2019).

Nos animais, a umidade exerce influência sobre a capacidade de perda de calor do animal exposto a elevadas temperaturas, pois quando esta atinge valores próximos a 72%, os bovinos perdem cerca de 20% da eficiência evaporativa (cutânea) e respiratória no que tange à perda de calor (BERMAN et al., 2016). Como mecanismo adaptativo inversamente proporcional ao calor, estes animais reduzem o consumo de matéria seca entre 3 a 10%, a fim de manter a homeotermia (NRC, 1981).

À vista disso, compreender a interação entre clima e animal torna-se imprescindível, assim como estabelecer critérios de classificação de ambientes, uma vez que na ausência de estresse térmico não há perda em rendimento e declínio na produção, para tal utiliza-se a escala térmica de ITU, onde, por via de regra, valores inferiores a 70 são considerados “confortáveis”, de 75 a 78 “estressantes” e acima de 78 impossibilita os animais de manterem a homeotermia (LEITE et al., 2012). Em condições tropicais, como no Brasil, onde

a temperatura ambiente facilmente ultrapassa os 26°C, é habitual confrontar-se com valores superiores a 75 na escala de ITU (SOUZA et al., 2010).

2.3 Comportamento de amamentação

Considera-se a fase de cria dos bovinos como um ponto essencial no sucesso da produção, uma vez que está atrelada às maiores taxas de mortalidade dos animais (NASCIMENTO et al., 2017), chegando até 75% de perdas logo no primeiro ano de vida (PRESTES; LANDIM-ALVARENGA, 2006), o que exige um melhor cuidado às adversidades susceptíveis aos bezerros no período pós-parto (PARANHOS DA COSTA; SILVA, 2011).

A primeira amamentação, ou colostragem, é a ação mais importante a realizar-se neste período (SALLES, 2011), uma vez que, os bezerros neonatos nascem agamaglobulinêmicos, ou seja, desprovidos de imunoglobulinas, as quais são adquiridas apenas na primeira amamentação, e são de extrema relevância na ação contra patógenos existentes no ambiente extrauterino (PARANHOS DA COSTA et al., 2007; CAIXETA; CARMO, 2020).

Neste sentindo, e levando em consideração que os bovinos desenvolvem comportamento gregário, admite-se maior importância à relação materno-filial, que compreende todas as atividades da matriz com sua cria, incluindo a fase de amamentação, o que afeta diretamente a taxa de sobrevivência e o desenvolvimento de bezerros (SOUZA-CONDE et al., 2015; SWAIN et al., 2015).

São muitos os fatores que podem influenciar o comportamento de amamentação, entre eles destacam-se as influências genéticas e ambientais (Day et al., 1987). O ambiente de criação deve possibilitar a expressão do potencial genético dos animais em sua totalidade, visando o aumento de produtividade (PEREIRA; OLIVEIRA, 2020).

Entre 30 e 60 dias de idade, os bezerros tendem a aumentar a duração de cada mamada, estabilizando-se aos 120 dias e reduzindo drasticamente ao aproximar-se do desmame. Segundo Paranhos da Costa et al. (2006), a frequência média de amamentação de bezerros é de três a cinco vezes por dia.

Já a duração está entre oito a onze minutos por ato de mamada, totalizando cerca de 30-60 minutos diários de amamentação.

A amamentação apresenta papel fundamental na produção de carne, uma vez que contribui com até 30% de variação no peso de bezerros pós-desmama, e com até 19% na diferença total do peso aos 365 dias de idade (ELER, 1989), demandando assim maior atenção à fase de cria com ênfase no desempenho de bovinos de corte.

2.4 Índice de Kleiber

As características de eficiência alimentar estão diretamente associadas com a lucratividade e sustentabilidade da bovinocultura de corte, por isso, preconiza-se a seleção de animais mais eficientes na conversão alimentar e qualidade de produtos cárneos, incrementando o lucro do produtor (FERNANDES et al., 2014).

Neste sentido, mensurar os fatores que influenciam no desenvolvimento dos animais torna-se fundamental na criação de bezerros (FIIRST et al., 2020), entretanto, as taxas de conversão alimentar são de difícil determinação, principalmente em sistemas extensivos. O fato do índice de ganho de peso estar intimamente relacionado aos níveis de eficiência (CAMERON, 1998), e ser uma variável de fácil mensuração, desenvolveu-se o Índice de Kleiber (IK) como alternativa aos cálculos de conversão alimentar (KLEIBER, 1936).

O Índice de Kleiber, proposto por Kleiber (1936), trata-se de um índice de eficiência alimentar capaz de identificar animais com alta eficiência de crescimento relativo ao tamanho corporal, sem a necessidade de mensuração individual de consumo (GOMES et al., 2012).

Neste caso, por estar altamente correlacionada com o ganho médio diário e taxa de crescimento, leva-se em consideração apenas o ganho de peso em relação ao kg de peso metabólico, ou seja, se há elevação no ganho de peso para um mesmo peso metabólico, denota maior crescimento sem elevar os níveis de energia de mantença pelo animal (KLEIBER, 1961).

O cálculo é realizado da seguinte forma: $IK=GPD/PV^{0,75}$, em que GPD é o ganho de peso diário, $PV^{0,75}$ é o peso vivo metabólico durante o período de

crescimento usado na avaliação. Para o IK, valores altos são mais favoráveis para a eficiência alimentar, o que indica crescimento corporal obtido sem aumento da exigência energética de mantença (ARCHER et al., 1999).

Embora esta não seja precisamente uma medida de eficiência alimentar, pode ser utilizada como alternativa às predições convencionais nos casos em que não há alteração na dieta durante o período experimental (BERRY; CROWLEY, 2013).

3. OBJETIVOS

3.1 Objetivo Geral

Determinar o efeito do estresse térmico sobre o comportamento de amamentação e desempenho por Índice de Kleiber (IK) de bezerros de três diferentes grupos genéticos, em ambientes de pastejo com e sem a presença de sombreamento.

3.2 Objetivos Específicos

Determinar o efeito do ambiente sobre o estresse térmico através de medidas de temperatura corpórea superficial (TCS) e equação de índice de Temperatura e Umidade (ITU) entre bezerros da raça Nelore e cruzados ½ Nelore x ½ Angus e ½ Nelore x ½ Pantaneiro. Avaliar a influência do ambiente sobre a frequência e duração de amamentação dos bezerros. E, por fim, mensurar o ganho de peso e eficiência por Índice de Kleiber (IK) dos animais, bem como a correlação entre todas as variáveis.

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CAPÍTULO 2 - COMPORTAMENTO DE AMAMENTAÇÃO, CRESCIMENTO E ÍNDICE DE KLEIBER EM BEZERROS DE DIFERENTES GRUPOS GENÉTICOS EM CONDIÇÕES DE SOMBREAMENTO

CHAPTER 2 - BREASTFEEDING BEHAVIOR, GROWTH AND KLEIBER INDEX IN CALVES OF DIFFERENT GENETIC GROUPS IN SHADING CONDITIONS

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Resumo

Objetivou-se avaliar o efeito do estresse térmico sobre o comportamento de amamentação e desempenho de bezerros Nelore (NE), e cruzados ½ Nelore x ½ Angus (NA) e ½ Nelore x ½ Pantaneiro (NP), em ambientes providos ou não de sombreamento natural, através das equações do Índice de Temperatura e Umidade (ITU) e Índice de Kleiber (IK). Foram avaliados 25 animais, a partir dos 30 dias de idade distribuídos em: 9 bezerros NE, 8 NA e 8 NP, das 7 às 17 horas, em ambientes sombreado e a pleno sol. Foram coletados dados de número de atos de mamar por dia (NM), duração média da mamada (DMM), temperatura corpórea superficial (TCS), ITU, ganho de peso e IK. O ITU apresentou diferença ($P<0,05$) entre os ambientes, sendo inferior no espaço sombreado. Animais NA apresentaram maior TCS (33,00±2.31), diferindo ($P<0,05$) de NE (31,71±1,65) e NP (31,38±1,99), e menor ($P<0,05$) atividade de amamentação, sugerindo redução do conforto térmico, entretanto, seu ganho de peso e IK foram superiores, diferindo ($P<0,05$) dos demais grupos genéticos. Já animais NE e NP não diferiram ($P>0,05$) em ganho de peso e IK. Portanto, os resultados sugerem que os bezerros NA, apesar do maior desconforto térmico, utilizaram de mecanismos de termólise para manterem ganho de peso e eficiência alimentar (IK) maiores que os demais grupos. O NP apresentou maior adaptabilidade térmica, além de ganho de peso e IK semelhantes aos animais NE.

Palavras-chave: Adaptação, Bovinocultura, Desempenho, Eficiência Alimentar, Índice de Temperatura e Umidade, Pantanal.

Abstract

The objective of this study was to evaluate the effect of thermal stress on the breastfeeding behavior and performance of Nellore calves (NE), ½ Nellore x ½ Angus (NA) and ½ Nellore x ½ Pantaneiro (NP) crossed, in environments with natural shading or not, through the equations of Temperature and Humidity Index (THI) and Kleiber Index (KI). Twenty-five animals were

evaluated from 30 days of age: 9 NE calves, 8 NA and 8 NP crossed, from 7 to 17 hours, in shaded and in full sun environments. Data on the number of feeding acts per day (NFA), mean duration of feeding (MDF), body surface temperature (BST), THI, weight gain and KI were collected. The THI showed a difference (P<0,05) between the environments, being inferior in the shaded one. NA animals presented higher BST (33,00±2.31), differing (P<0,05) from NE and NP, and lower (P<0,05) breastfeeding activity, suggesting a reduction in thermal comfort, however their weight gain and KI were higher, differing (P<0,05) from the other genetic groups. Therefore, the results suggest that NA calves, despite the greater thermal discomfort, used thermolysis mechanisms to maintain weight gain and feed efficiency (KI) higher than the other groups. NP animals showed higher thermal adaptability to the environment, in addition to weight gain and KI similar to NE animals.

Keywords: Adaptation, Cattle breeding, Food Efficiency, Pantanal, Performance, Temperature and Humidity Index.

1. INTRODUÇÃO

A bovinocultura de corte praticada em sistema extensivo, é a atividade com maior importância econômica no Pantanal brasileiro, bioma caracterizado por altas temperaturas e umidade durante todo o ano (OLIVEIRA et al., 2016), atingindo no ano de 2020 médias de 25,6ºC e 68,59%, para estes parâmetros, respectivamente (INMET, 2021).

Isso torna primordial a identificação e seleção de raças que ali serão criadas, de modo a reduzir os efeitos da interação genótipo-ambiente (OLIVEIRA et al., 2017), uma vez que o estresse térmico pode afetar funções bioenergéticas (BROWN-BRANDL, 2018), reduzindo o consumo de alimentos, crescimento, eficiência alimentar, imunidade e, consequentemente, aumento da taxa de mortalidade e prejuízos econômicos (ST-PIERRE et al., 2003).

Uma alternativa é o uso de raças de perfil adaptativo aos fatores climáticos do Pantanal, como o caso da raça Pantaneiro, naturalizada deste bioma (REZENDE et al., 2014) devido a sua elevada rusticidade e ínferas exigências nutricionais, a qual atualmente sofre risco de extinção, ou a raça zebuína Nelore, já amplamente utilizada nesta região, e que respondem bem fisiologicamente às intempéries (BARBOSA et al., 2017).

Na busca por melhores índices produtivos e qualitativos de carcaça, alguns criadores têm iniciado a inclusão da raça Angus em sistemas de cruzamento, entretanto, por ser de origem europeia, é considerada inferior em controle do estresse térmico (BATTISTELLI et al., 2013). Logo, torna-se importante o uso de cruzamentos entre raças no sentido de amenizar os impactos do estresse térmico e manter a qualidade de produtos cárneos, de modo a não serem necessárias intervenções gerenciais e a evitar prejuízos econômicos (SCHOLTZ et al., 2011).

Apesar de considerar-se que animais Angus possuem melhores índices produtivos quando comparados aos indivíduos das raças Nelore e Pantaneiro (FERREIRA et al., 2019), em um ambiente como o Pantanal, são escassos os estudos que deem subsídio a hipótese de que o estresse térmico possa influenciar na redução da superioridade em desempenho de mestiços ½ Nelore x ½ Angus em comparação com animais Nelores ou mestiços ½ Nelore x ½ Pantaneiro.

Neste propósito, é essencial o desenvolvimento de métodos de mensuração eficazes de estresse térmico como uma ferramenta valiosa de potencialização da produtividade animal, entre estes destaca-se o Índice de Temperatura e Umidade (ITU), o qual classifica o estresse térmico através da combinação de dados simples e acurados como a temperatura e umidade locais (THOM, 1959).

Simultaneamente a esta avaliação, levando em consideração que a amamentação contribui com até 19% na diferença total do peso aos 365 dias de idade, e que esta é diretamente influenciada por fatores ambientais, como as altas temperaturas, faz-se necessário a mensuração da temperatura corpórea superficial e sua relação com o comportamento de amamentação, de modo a aferir o efeito do estresse térmico sobre o desempenho desses animais.

Neste sentido, aliado aos métodos avaliativos descritos anteriormente, tem-se a necessidade da utilização de um índice de mensuração de eficiência alimentar, entretanto, a maior parte dessas estimativas depende de dados de consumo individual, o que inviabiliza a coleta em um número expressivo de animais (ARTHUR et al., 2004). Como alternativa, Kleiber (1936), desenvolveu uma equação chamada Índice de Kleiber (IK), que é capaz de predizer o desempenho de bovinos através do ganho de peso médio diário (GMD) e o peso vivo metabólico, não requerendo medidas individuais de ingesta (KLEIBER, 1936).

Assim, objetivou-se avaliar o efeito das condições climáticas da região do Pantanal, com variações de sombreamento natural e sob pleno sol, sobre o comportamento de mamada e Índice de Kleiber de bezerros Nelore, ½ Nelore x ½ Angus e ½ Nelore x ½ Pantaneiro.

2. MATERIAL E MÉTODOS

O estudo foi conduzido entre agosto de 2019 e maio de 2020, no centro experimental de Bovinocultura de Corte da Universidade Estadual de Mato Grosso do Sul, Campus de Aquidauana-MS, região do Alto Pantanal, situado em latitude 20º30’S, longitude 55º50’W (Brasil), e altitude de 167 metros. O clima da região, segundo a classificação de Köppen, é Tropical Savana com inverno seco, e verão com possibilidades de temperaturas superiores à 35ºC.

A Comissão de Ética no Uso de Animais (CEUA-UEMS) certificou a utilização dos mesmos de acordo com o protocolo nº 029/2019. Foram avaliados 25 bezerros contemporâneos, a partir dos 30 dias de idade, filhos de vacas Nelore, com intervalo de nascimento de no máximo 10 dias, distribuídos em três grupamentos genéticos: Nelore (NE; n=9); cruzado ½ Nelore x ½ Angus (NA; n=8); e cruzado ½ Nelore x ½ Pantaneiro (NP; n=8).

2.1 Coleta de dados

Todos os animais envolvidos na pesquisa (matrizes e bezerros) foram previamente identificados com números marcados em seus flancos. Foram realizadas 5 (cinco) coletas

amostrais, cada qual consistindo em 10 horas de observação, iniciando às 7 horas da manhã e finalizando às 17 horas da tarde. Os ambientes experimentados foram dois piquetes de 10 hectares cada, ambos formados pela forrageira *Urochloa brizantha*, com a dessemelhança da presença de sombreamento natural de árvores nativas em um, e outro desprovido de condições de sombreamento seja natural ou artificial.

Cada período amostral resumiu-se em três dias consecutivos, sendo o primeiro destinado apenas a pesagem dos animais, o segundo às observações no ambiente desprovido de sombreamento, e o terceiro dia em coletas no ambiente sombreado. Os animais eram transferidos de um ambiente para outro ao final do segundo dia, para que o manejo não afetasse as coletas de comportamento do dia subsequente.

As informações apuradas a respeito do comportamento de amamentação consistiam em número de atos de mamar por dia (NM) e duração média da mamada (DMM), segundo a metodologia de Castanheira (2013).

Sobre as mensurações de conforto térmico, foram coletadas durante todo o período amostral, a cada hora, a temperatura corpórea superficial (TCS) da região das costelas dos animais, com o uso de um termômetro digital infravermelho laser sensorial (marca *Btuty*, modelo HW600, com escala de -50 a 600°C, China); e a temperatura ambiente (TA), através de um Termo-higrômetro (marca *Icoterm*, modelo Analógico Bulbo Seco e Úmido, com escala de -10 a 50°C, Porto Alegre-RS, Brasil). Ademais, utilizou-se o banco de informações da Estação de Sistema de Navegação Global por Satélite - GNSS, implantada na Universidade Estadual de Mato Grosso do Sul (UEMS) - Campus de Aquidauana-MS, para extrair as informações de temperatura (TºC) e umidade relativa (UR) do ar, utilizados na predição do índice de temperatura e umidade (ITU).

Em equivalência ao cálculo de ITU, utilizou-se a equação reportada por Thom (1959), sendo: ITU=(0,8 x TºC+(UR(%)/100) x (TºC-14,4)+46,4), em que: TºC se dá pela temperatura ambiente em graus Celsius, e UR pela umidade relativa do ar. A interpretação do cálculo de ITU baseou-se no modelo definido por Armstrong (1994), em que considera valores entre 72 a 78 como potencial estresse térmico ameno ou brando, 79 a 88 como potencial estresse moderado e 89 a 98 como potencial estresse severo. Este cálculo foi realizado em todos os dias do período experimental, a cada hora, durante todo intervalo de coletas (7 horas da manhã às 17 horas da tarde).

Em relação aos dados de desempenho, foram estimados os índices de ganho médio diário dos animais utilizando os dados de pesagem e a seguinte equação: GMD=(PF-PI)/N, onde PF é o peso final, PI é o peso inicial e N é o número de dias do período. Além disso, foi

determinado o Índice de Kleiber, através da equação: $IK=(GMD/P^{0.75})*100$ (KLEIBER, 1936), onde GMD é o ganho médio diário e $P^{0.75}$ é o peso metabólico do animal.

2.2 Análises estatísticas

Foi utilizado o software R (2018) e Past (Hammer et al., 2001) para análise estatística dos dados. Inicialmente, foi avaliada a distribuição do ITU e as diferenças para seus valores entre a condição de sombreamento ou pleno sol, bem como efeitos do período do dia e coleta. Posteriormente, foi testada para a TCS a hipótese de igualdade em um modelo que levou em consideração os efeitos do período do dia, condição do ambiente (provido ou desprovido de sombreamento), grupamento genético e dia da coleta, bem como a interação entre estes fatores.

Realizou-se a análise de regressão por grupo genético considerando o ITU e TCS, e desconsiderando as condições ambientais (sombreamento e pleno sol), haja vista que as diferenças de ITU entre estas condições não foram suficientes para alterar a classificação de estresse moderado, de acordo com a classificação de Armstrong (1994) adotada por este trabalho.

Adicionalmente, calculou-se a MANOVA com intuito de verificar diferenças entre os grupos genéticos, considerando simultaneamente as variáveis indicativas de estresse térmico (ITU e TCS) e o comportamento na amamentação. Posteriormente, realizou-se análise canônica, onde o número de CAN extraído foi definido de acordo com o critério de variância, ou seja, o ponto de inflexão no gráfico da curva de autovalores, estes foram usados na elaboração de um gráfico biplot onde cada linha representa um ponto Gn com uma coordenada (gi1, gi2), i = 1, • • •, n, e cada coluna um vetor começa na origem até o ponto Hm com uma coordenada (hj1, hj2), j = 1, • • •, p, ou seja, os vetores representam as variáveis antes mencionadas, e os pontos representam os grupos genéticos.

3. RESULTADOS

O ITU apresentou valores oscilando entre 76,46 a 90,69 (Figura 1-A), com diferença significativa ($P<0,05$) entre os ambientes sombreado (83,06±1,75) e de pleno sol (85,37±1,89) (Figura 1-B). Diferenças também foram observadas ($P<0,05$) para o horário do dia e entre os períodos amostrais.

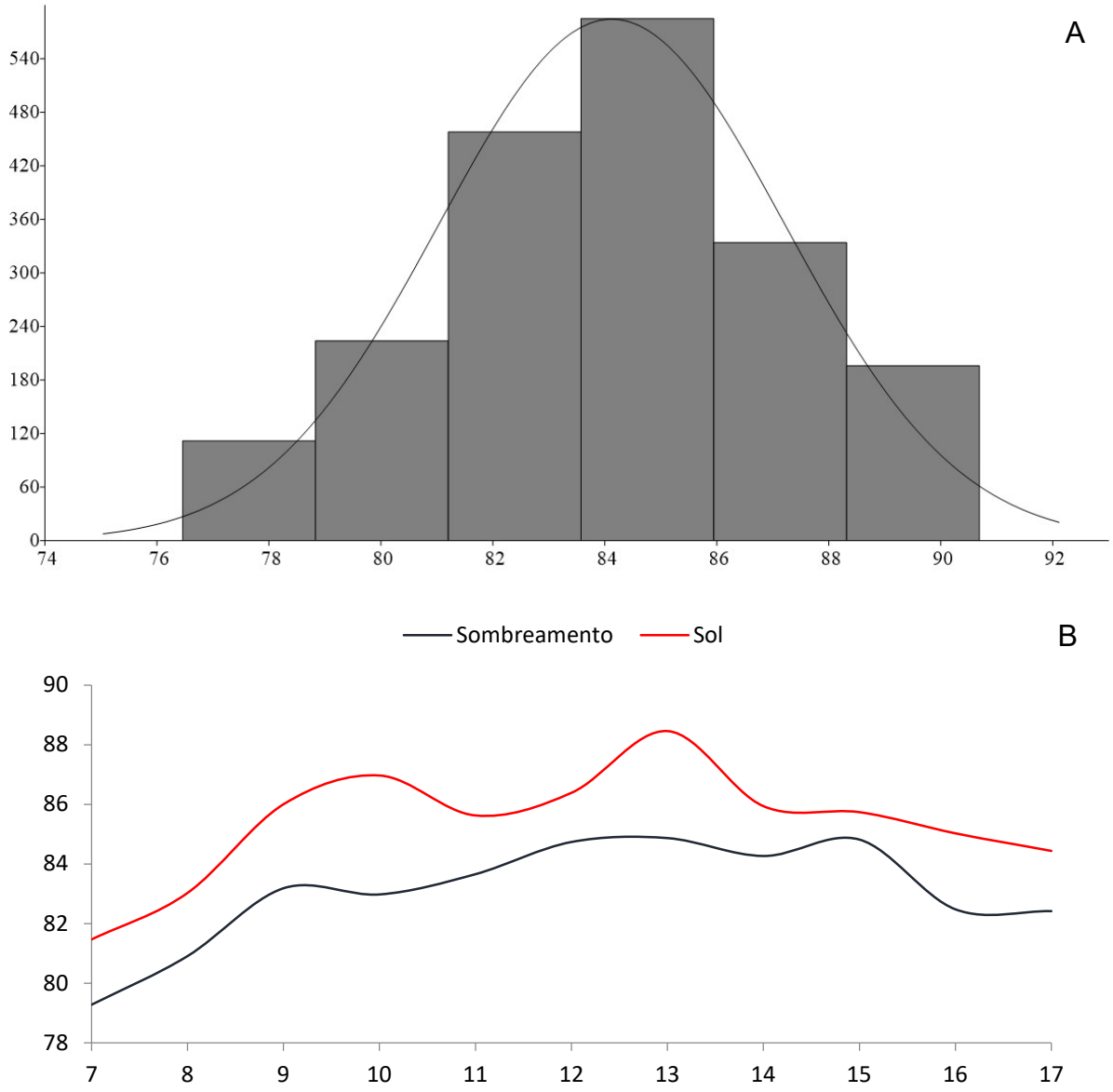
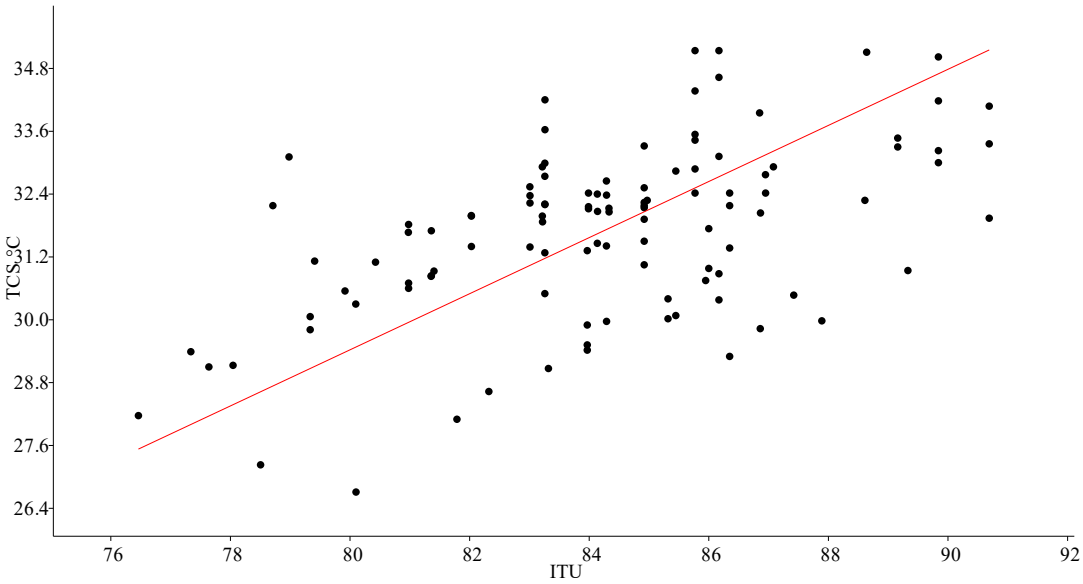


Figura 1. Distribuição dos valores de ITU durante todo o período experimental (A) e entre os horários de coleta (B) em ambientes providos ou não de sombreamento. Valores de P = Coleta (P<0,05); Horário (P<0,05); Ambiente (P<0,05).

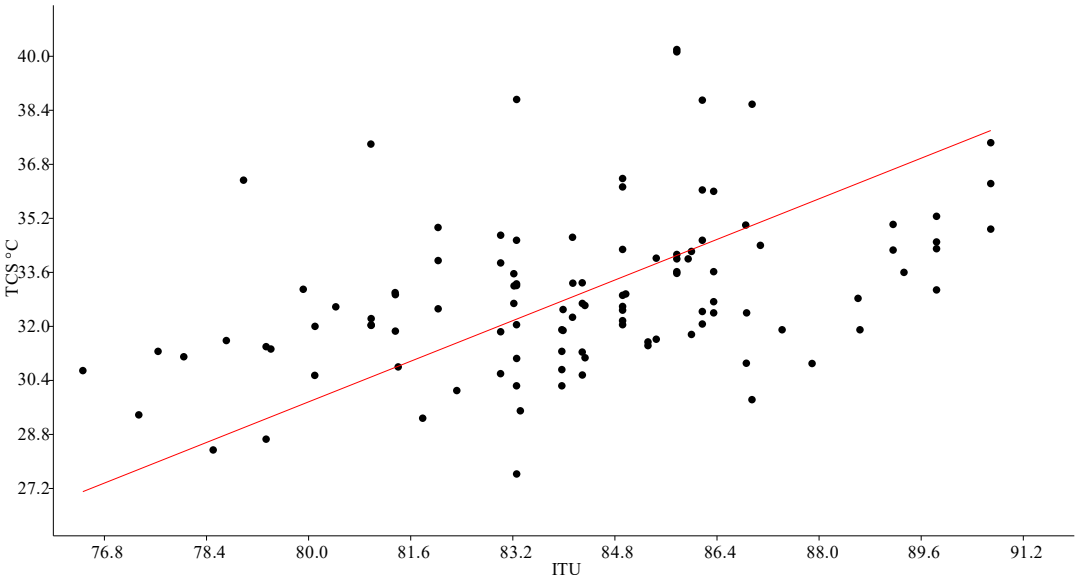
Todos os efeitos considerados no modelo influenciam (P<0,05) nos resultados do TCS, sendo o efeito com maior acúmulo de variância o horário do dia, seguido do grupo genético. Animais NA apresentaram maior TCS (33,00±2,31), sendo diferente significativamente do NE (31,71±1,65) e do NP (31,38±1,99).

Todavia, o ITU apresentou relação positiva de magnitude baixa (P<0,05) com a TCS, com valores dos coeficientes de determinação de 0,28, 0,16 e 0,19 respectivamente para o NE (Figura 2-A), NA (Figura 2-B) e NP (Figura 2-C).

A


TCS, °C
34.8
33.6
32.4
31.2
30.0
28.8
27.6
26.4
76
78
80
82
84
86
88
90
92
ITU

B


TCS °C
40.0
38.4
36.8
35.2
33.6
32.0
30.4
28.8
27.2
76.8
78.4
80.0
81.6
83.2
84.8
86.4
88.0
89.6
91.2
ITU

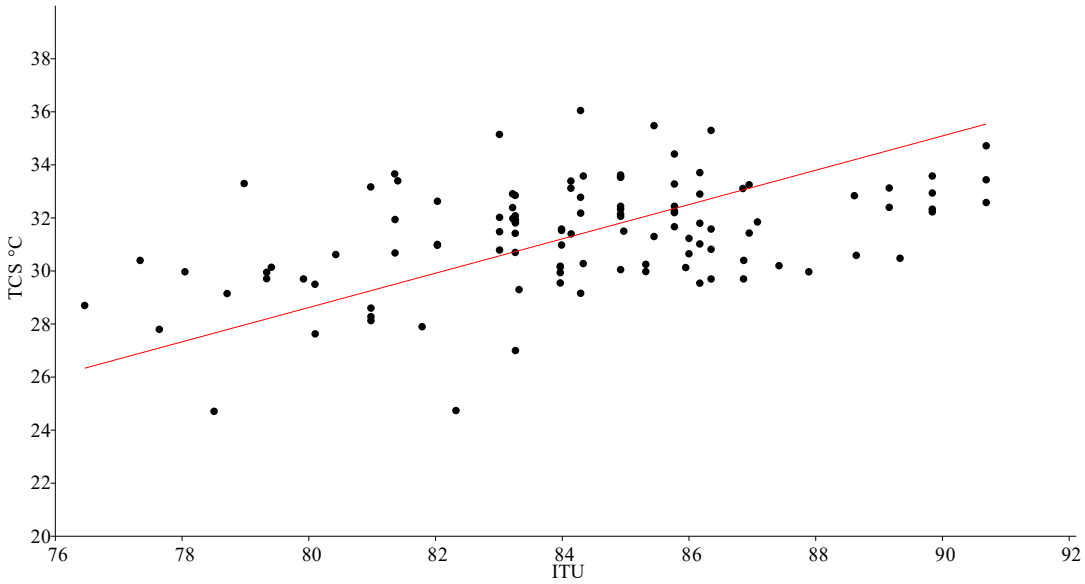


Figura 2. Relação do Índice de Temperatura e Umidade (ITU) e temperatura corpórea superficial (TCS) (°C) em bezerros NE (A), NA (B) e NP (C).

Foram observadas diferenças significativas (P<0,05) entre NA em relação ao NP quando consideradas simultaneamente (MANOVA) as variáveis indicativas de estresse térmico (ITU e TCS) e comportamento de amamentação. O NE não diferiu (P>0,05) dos demais, com grande variação de respostas entre seus indivíduos. O NA apresentou maior TCS conjuntamente à menor atividade de amamentação (Figura 3).

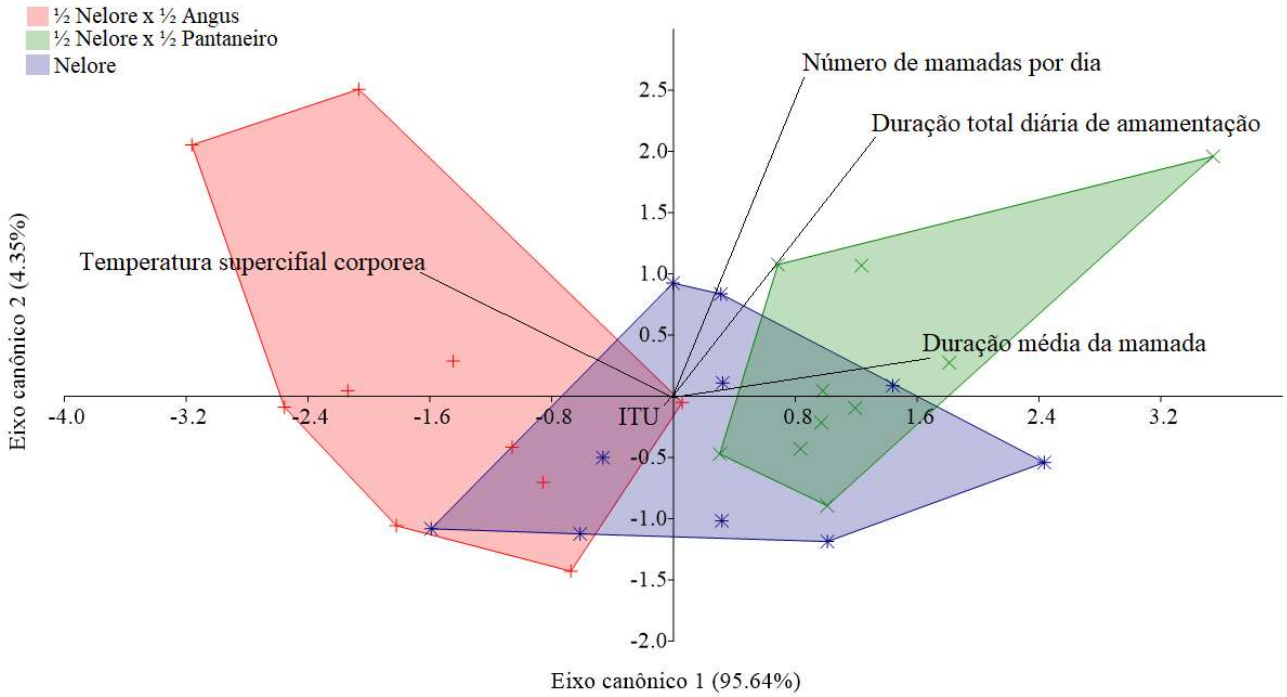
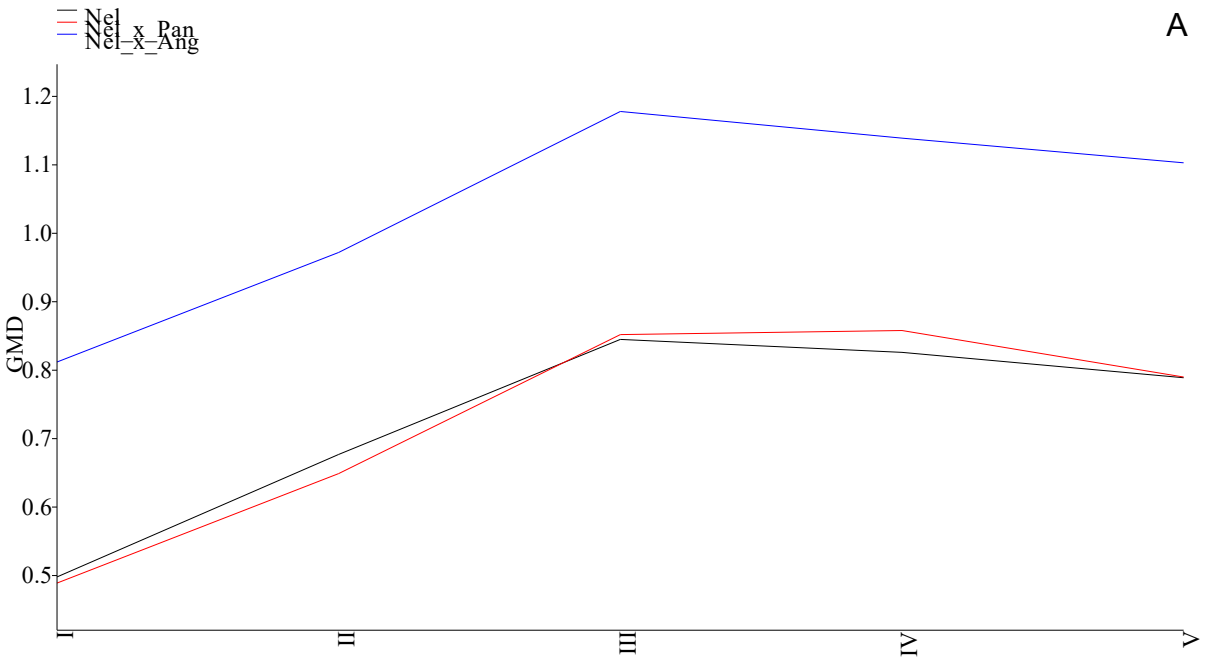


Figura 3. Biplot considerando o Índice de Temperatura e Umidade (ITU) e temperatura corpórea superficial (TCS) (ºC) e comportamento durante amamentação para o NE, NA e NP.

O IK e GMD foram maiores para o NA, apresentando diferenças significativas (P<0,05) (Figura 4-A). Entre NE e NP, não houveram diferenças significativas (P>0,05).



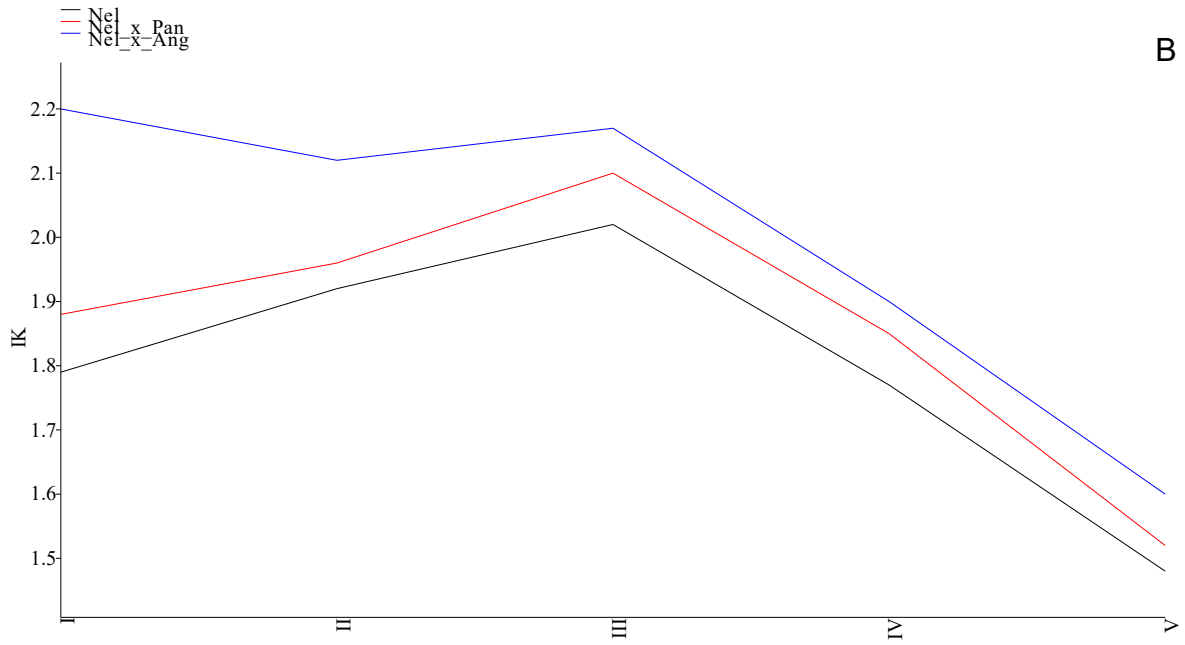


Figura 4. Ganho de peso (A) e índice de Kleiber (B) durante período amostral (I a V).

4. DISCUSSÃO

Indicadores de estresse térmico

Os níveis de ITU encontrados neste trabalho indicaram existência de estresse térmico que, segundo a metodologia de Armstrong (1994), variam entre desconforto ameno a severo. Ao comparar a condição de cada ambiente, provido ou não de sombreamento, a área sombreada propiciou níveis significativamente inferiores de ITU. Esta diferença é justificada pela condição de troca térmica auxiliada pelas árvores, que favorece a atuação de mecanismos de redução de estresse térmico (BAÊTA; SOUZA, 2010). O mesmo foi relatado anteriormente em diversos estudos (MORALES et al., 2017; JOSET et al., 2018; ARANHA et al., 2019; BOSI et al., 2020), os quais afirmam que oportunizar áreas sombreadas à criação de bovinos de corte reduz os níveis de ITU entre 3,7 (KARVATTE et al., 2016) a 8% (ALVES et al., 2019).

Neste trabalho, apesar da esperada redução nos níveis de ITU no local sombreado, a diferença entre os ambientes foi de apenas 2,83%, não sendo suficiente para alterar a classificação de estresse moderado para estresse ameno, ou inexistente. Isso pode ser justificado pelas altas temperaturas anuais características da região pantaneira, aliado ao fato das estações em que foram desenvolvidas o estudo serem qualificadas como o período mais crítico do ano em termos de conforto térmico (NASCIMENTO et al., 2017).

O horário do dia interferiu diretamente nos níveis de ITU, sendo o momento mais crítico por volta das 13 horas da tarde, assim como nos trabalhos de Moraes Júnior et al. (2010), Cavalcante et al. (2017) e Souza et al. (2017), os quais também destacaram maior desconforto térmico no período vespertino. Este tipo de situação pode influenciar não apenas o comportamento dos animais, como também suas condições fisiológicas, os quais passam a mobilizar energia para a termorregulação em detrimento do ganho de peso (SANTOS; CABRAL, 2021).

Ainda a respeito dos índices de estresse térmico, corroborando com Pantoja et al. (2018), os níveis de TCS encontrados apresentaram correlação positiva com os dados de ITU, exprimindo o fato de que quanto maior o índice de temperatura e umidade, mais elevada será a temperatura corpórea superficial; assim como a TCS encontrada neste trabalho acompanha sincronicamente, e significativamente a variação da temperatura ao longo do dia.

Animais oriundos do cruzamento NA apresentaram maior TCS, em resposta ao aumento proporcional nos níveis de ITU, indicando que exemplares de padrões raciais NA tendem a sofrer mais com altas temperaturas em comparação com cruzados NP e NE.

Isto pode se justificar na diferença de pelame entre os grupos genéticos pois, segundo Silva (2000), em regiões de climas quentes, animais com características de pelos menos espessos e menos densos são mais interessantes na facilitação da termorregulação, pois favorecem a perda de calor por convecção. Sabe-se que mestiços NA apresentam maior comprimento, número e densidade de pelos que animais NE (RIBEIRO et al., 2008), além disso, animais NE apresentam maior ângulo de inclinação do pelo em relação à epiderme (MÜLLER, 1989), o que favorece a adaptação destes ao clima tropical comparativamente aos mestiços NA.

Outra característica adaptativa do grupo NE é sua epiderme altamente pigmentada em combinação com pelame claro, uma consequência da seleção natural de espécies adaptadas às elevadas temperaturas, que visa proteger os tecidos da ação da radiação ultravioleta (SILVA et al., 2003). Em trabalho de Aranha et al. (2019), não houve diferença de desempenho em animais da raça Nelore entre ambientes providos e desprovidos de conforto térmico, confirmando a aclimatação da raça ao clima tropical. Ademais, de acordo com Müller (1989), zebuínos possuem maior número de glândulas sudoríparas que os taurinos, ou seja, maior facilidade em eliminar calor pelo suor, além disso, são providos de alelos relacionados à termotolerância e, consequentemente, maior capacidade adaptativa (HANSEN, 2004).

Todavia, animais taurinos também são capazes de desenvolver habilidades adaptativas, como o caso da raça naturalizada Pantaneiro, que se adaptaram por meio da seleção natural às condições alagadas e climáticas do Pantanal pelos últimos três séculos (McMANUS et al.,

2009). Segundo Dani e Oliveira (2013) e Barbosa et al. (2014), estes animais conseguem manter a homeostase, a normalidade de parâmetros fisiológicos e atividades comportamentais, como a demanda de menor consumo de água e maior interação social mesmo em períodos críticos de temperatura (LIMA et al., 2019). Estes traços podem ser justificados fisiologicamente à espessura de pelame e elevado número de glândulas sudoríparas, essenciais na dissipação de calor (MAZZA et al., 1992; BIANCHINI et al., 2006).

Ainda com relação à TCS, animais NE não diferiram significativamente de cruzados NP. Essa similaridade demonstra que ambas as raças apresentam maior habilidade adaptativa ao clima do bioma Pantanal.

Efeito do estresse térmico no comportamento de amamentação e Índice de Kleiber entre os grupos genéticos

Simultaneamente às variáveis de TCS e ITU, notou-se o nível de influência do aumento da temperatura (ambiente e superficial corpórea) sobre o comportamento de amamentação dos bezerros, a exemplo dos cruzados NA, animais com maiores índices de TCS, que obtiveram menor atividade de amamentação. Em contrapartida, os NE e os cruzados NP, de menores TCS, apresentaram mais ações para amamentação, denotando maior resistência e adaptabilidade às altas temperaturas. Para Das et al. (2000), animais zebuínos apresentam 0,6 vezes mais de frequência e 9,4 minutos a mais de duração de mamada em comparação com mestiços ½ zebuíno x ½ taurino.

Com relação ao ganho de peso, para Soberon et al. (2012), é durante o período de amamentação que os efeitos genéticos relacionados a esta variável são expressos, sendo os mesmos reflexos da qualidade e quantidade da ingestão de leite até o desmame. Entretanto, contrariando o dado anterior, independente do maior estresse térmico demonstrado pelos animais NA, o qual refletiu em menores eventos de mamadas, o GMD desses animais foi superior em comparação com animais NE e NP. Isto é justificado por sua maior eficiência (IK), a qual também foi significativamente superior aos demais grupos genéticos, que indica uma maior diluição das necessidades de mantenças.

Já entre os grupos NE e NP, não houveram diferenças significativas para GMD e IK, o que denota uma semelhança não apenas adaptativa, mas também em desempenho produtivo entre essas raças. Isso demonstra que, a vantagem da introdução de genes pantaneiros no rebanho vai além da questão adaptativa, visto que, com relação às características produtivas, o cruzamento NP se mostra semelhante em ganho de peso aos animais Nelore.

5. CONCLUSÃO

Os bezerros cruzados ½ Nelore x ½ Angus sofreram maior desconforto térmico em comparação com bezerros Nelore e cruzados ½ Nelore x ½ Pantaneiro, apresentando maiores índices de temperatura corpórea superficial e menor atividade de amamentação. Em contrapartida, apesar do ambiente desfavorável, os animais ½ Nelore x ½ Angus utilizaram de mecanismos de termólise para resistir ao desconforto térmico, apresentando maior ganho de peso e eficiência alimentar, mensurada pelo Índice de Kleiber, em comparação aos demais grupamentos genéticos. Os bezerros ½ Nelore x ½ Pantaneiro apresentaram maior adaptabilidade térmica ao ambiente, além de ganho de peso e Índice de Kleiber semelhantes aos animais Nelore.

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CAPÍTULO 3 - CONSIDERAÇÕES FINAIS

Animais taurinos apresentam melhores índices produtivos quando comparados às raças Nelore e mestiços ½ Nelore x ½ Pantaneiro, entretanto, melhores características adaptativas às intempéries da região do Pantanal, provêm da raça naturalizada Pantaneiro, a qual está sob o risco de extinção devido a escolha por raças mais produtivas, em detrimento de raças nativas.

Ademais, a vantagem da utilização do cruzamento ½ Nelore x ½ Pantaneiro não se dá apenas na adaptabilidade destes animais, uma vez que demonstraram semelhança em ganho de peso e índice de Kleiber aos animais Nelores. Isso denota a capacidade desta raça em equivaler-se, ou até mesmo superar, animais comumente utilizados nesta região.

Os cruzamentos entre raças vêm se tornando um procedimento cada vez mais comum e acessível ao produtor de bovinos de corte. A realidade climática do Pantanal exige a seleção de animais com capacidades adaptativas e habilidade termorregulatória superior, sem dispensar características produtivas como eficiência alimentar e ganho de peso, por isso a preservação de material genético é essencial na seleção de bovinos de corte.

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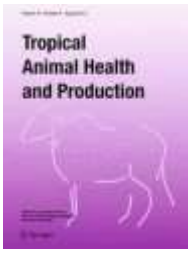
ANEXOS

ANEXO 1 - Normas para submissão: Revista Tropical Animal Health and Production



Tropical Animal Health and Production

Submission guidelines

Contents

Instructions for Authors

Authorship Policy

Authorship should incorporate and should be restricted to those who have contributed substantially to the work in one or more of the following categories:

• Conceived of or designed study

• Performed research

• Analyzed data

• Contributed new methods or models

• Wrote the paper

- It is the responsibility of the corresponding authors that the names, addresses and affiliations of all authors are correct and in the right order, that institutional approvals have been obtained and that all authors have seen and agreed to a submission. This includes single authorship papers where appropriate. If at all in doubt please double check with eg. Supervisors, line managers department heads etc.

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Manuscript Submission

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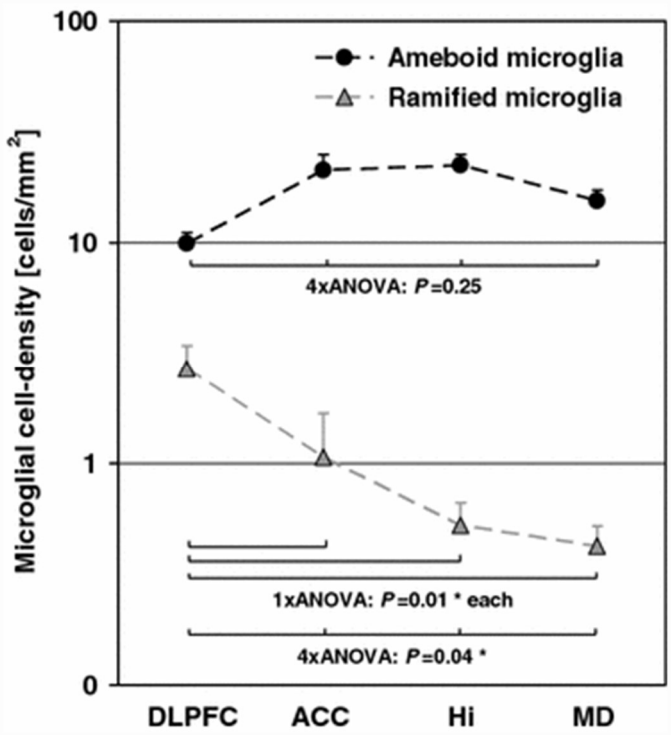

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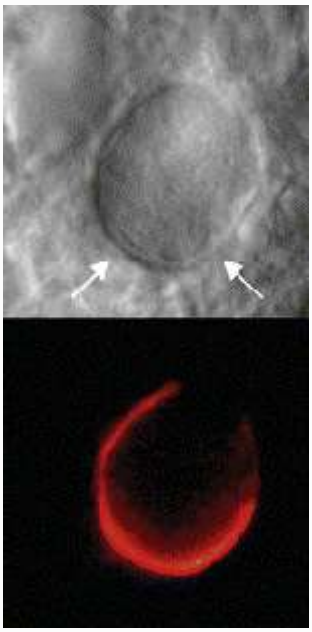

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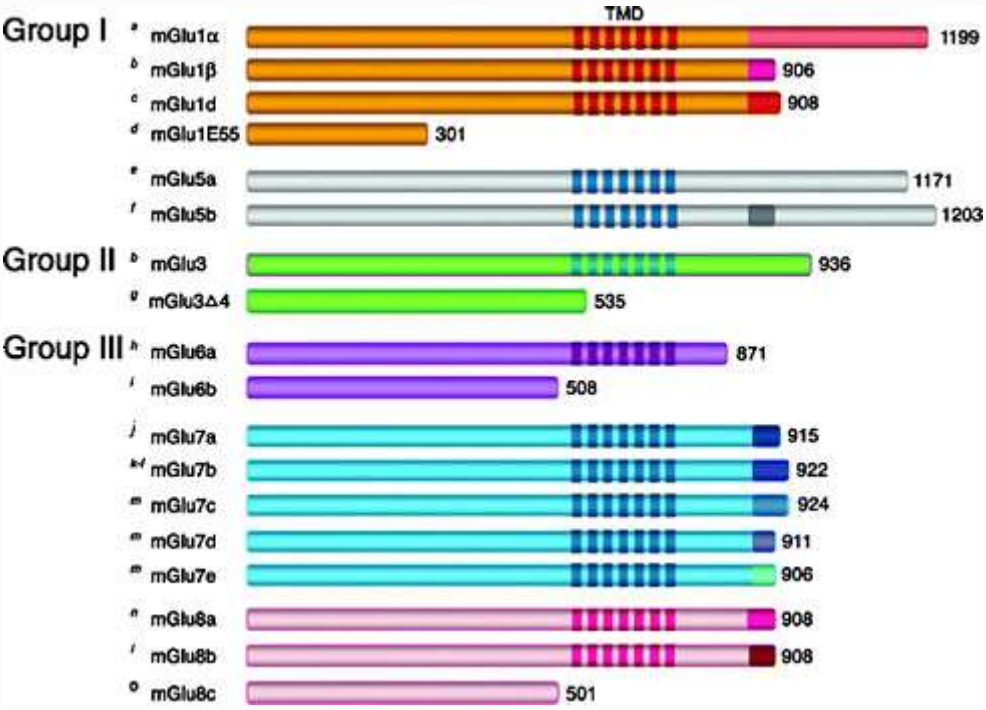

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The Editors reserve the right to reject manuscripts that do not comply with the above-mentioned guidelines. The author will be held responsible for false statements or failure to fulfill the above-mentioned guidelines.

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Competing Interests

Authors are requested to disclose interests that are directly or indirectly related to the work submitted for publication. Interests within the last 3 years of beginning the work (conducting the research and preparing the work for submission) should be reported. Interests outside the 3-year time frame must be disclosed if they could reasonably be perceived as influencing the submitted work. Disclosure of interests provides a complete and transparent process and helps readers form their own judgments of potential bias. This is not meant to imply that a financial relationship with an organization that sponsored the research or compensation received for consultancy work is inappropriate.

Editorial Board Members and Editors are required to declare any competing interests and may be excluded from the peer review process if a competing interest exists. In addition, they should exclude themselves from handling manuscripts in cases where there is a competing interest. This may include – but is not limited to – having previously published with one or more of the authors, and sharing the same institution as one or more of the authors. Where an Editor or Editorial Board Member is on the author list they must declare this in the competing interests section on the submitted manuscript. If they are an author or have any other competing interest regarding a specific manuscript, another Editor or member of the Editorial Board will be assigned to assume responsibility for overseeing peer review. These submissions are subject to the exact same review process as any other manuscript. Editorial Board Members are welcome to submit papers to the journal. These submissions are not given any priority over other manuscripts, and Editorial Board Member status has no bearing on editorial consideration.

Interests that should be considered and disclosed but are not limited to the following:

Funding: Research grants from funding agencies (please give the research funder and the grant number) and/or research support (including salaries, equipment, supplies, reimbursement for attending symposia, and other expenses) by organizations that may gain or lose financially through publication of this manuscript.

Employment: Recent (while engaged in the research project), present or anticipated employment by any organization that may gain or lose financially through publication of this manuscript. This includes multiple affiliations (if applicable).

Financial interests: Stocks or shares in companies (including holdings of spouse and/or children) that may gain or lose financially through publication of this manuscript; consultation fees or other forms of remuneration from organizations that may gain or lose financially; patents or patent applications whose value may be affected by publication of this manuscript.

It is difficult to specify a threshold at which a financial interest becomes significant, any such figure is necessarily arbitrary, so one possible practical guideline is the following: "Any undeclared financial interest that could embarrass the author were it to become publicly known after the work was published."

Non-financial interests: In addition, authors are requested to disclose interests that go beyond financial interests that could impart bias on the work submitted for publication such as professional interests, personal relationships or personal beliefs (amongst

others). Examples include, but are not limited to: position on editorial board, advisory board or board of directors or other type of management relationships; writing and/or consulting for educational purposes; expert witness; mentoring relations; and so forth.

Primary research articles require a disclosure statement. Review articles present an expert synthesis of evidence and may be treated as an authoritative work on a subject. Review articles therefore require a disclosure statement.Other article types such as editorials, book reviews, comments (amongst others) may, dependent on their content, require a disclosure statement. If you are unclear whether your article type requires a disclosure statement, please contact the Editor-in-Chief.

Please note that, in addition to the above requirements, funding information (given that funding is a potential competing interest (as mentioned above)) needs to be disclosed upon submission of the manuscript in the peer review system. This information will automatically be added to the Record of CrossMark, however it is **not added** to the manuscript itself. Under 'summary of requirements' (see below) funding information should be included in the '**Declarations**' section.

Summary of requirements

The above should be summarized in a statement and placed in a 'Declarations' section before the reference list under a heading of 'Funding' and/or 'Competing interests'. Other declarations include Ethics approval, Consent, Data, Material and/or Code availability and Authors' contribution statements.

Please see the various examples of wording below and revise/customize the sample statements according to your own needs.

When all authors have the same (or no) conflicts and/or funding it is sufficient to use one blanket statement.

Examples of statements to be used when funding has been received:

- Partial financial support was received from [...]
- The research leading to these results received funding from [...] under Grant Agreement No[...].
- This study was funded by [...]
- This work was supported by [...] (Grant numbers [...] and [...]

Examples of statements to be used when there is no funding:

- The authors did not receive support from any organization for the submitted work.
- No funding was received to assist with the preparation of this manuscript.
- No funding was received for conducting this study.
- No funds, grants, or other support was received.

Examples of statements to be used when there are interests to declare:

- **Financial interests:** Author A has received research support from Company A. Author B has received a speaker honorarium from Company Wand owns stock in Company X. Author C is consultant to company Y.

  Non-financial interests: Author C is an unpaid member of committee Z.

- **Financial interests:** The authors declare they have no financial interests.

  Non-financial interests: Author A is on the board of directors of Y and receives no compensation as member of the board of directors.

- **Financial interests:** Author A received a speaking fee from Y for Z. Author B receives a salary from association X. X where s/he is the Executive Director.

  Non-financial interests: none.

- **Financial interests:** Author A and B declare they have no financial interests. Author C has received speaker and consultant honoraria from Company M and Company N. Dr. C has received speaker honorarium and research funding from Company M and Company O. Author D has received travel support from Company O.

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Examples of statements to be used when authors have nothing to declare:

- The authors have no relevant financial or non-financial interests to disclose.
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- All authors certify that they have no affiliations with or involvement in any organization or entity with any financial interest or non-financial interest in the subject matter or materials discussed in this manuscript.

- The authors have no financial or proprietary interests in any material discussed in this article.

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Research involving human participants, their data or biological material

Ethics approval

When reporting a study that involved human participants, their data or biological material, authors should include a statement that confirms that the study was approved (or granted exemption) by the appropriate institutional and/or national research ethics committee (including the name of the ethics committee) and certify that the study was performed in accordance with the ethical standards as laid down in the 1964 Declaration of Helsinki and its later amendments or comparable ethical standards. If doubt exists whether the research was conducted in accordance with the 1964 Helsinki Declaration or comparable standards, the authors must explain the reasons for their approach, and demonstrate that an independent ethics committee or institutional review board explicitly approved the doubtful aspects of the study. If a study was granted exemption from requiring ethics approval, this should also be detailed in the manuscript (including the reasons for the exemption).

Retrospective ethics approval

If a study has not been granted ethics committee approval prior to commencing, retrospective ethics approval usually cannot be obtained and it may not be possible to consider the manuscript for peer review. The decision on whether to proceed to peer review in such cases is at the Editor's discretion.

Ethics approval for retrospective studies

Although retrospective studies are conducted on already available data or biological material (for which formal consent may not be needed or is difficult to obtain) ethics approval may be required dependent on the law and the national ethical guidelines of a country. Authors should check with their institution to make sure they are complying with the specific requirements of their country.

Ethics approval for case studies

Case reports require ethics approval. Most institutions will have specific policies on this subject. Authors should check with their institution to make sure they are complying with the specific requirements of their institution and seek ethics approval where needed. Authors should be aware to secure informed consent from the individual (or parent or guardian if the participant is a minor or incapable) See also section on **Informed Consent**.

Cell lines

If human cells are used, authors must declare in the manuscript: what cell lines were used by describing the source of the cell line, including when and from where it was obtained, whether the cell line has recently been authenticated and by what method. If cells were bought from a life science company the following need to be given in the manuscript: name of company (that provided the cells), cell type, number of cell line, and batch of cells.

It is recommended that authors check the NCBI database for misidentification and contamination of human cell lines. This step will alert authors to possible problems with the cell line and may save considerable time and effort.

Further information is available from the International Cell Line Authentication Committee (ICLAC).

Authors should include a statement that confirms that an institutional or independent ethics committee (including the name of the ethics committee) approved the study and that informed consent was obtained from the donor or next of kin.

Research Resource Identifiers (RRID)

Research Resource Identifiers (RRID) are persistent unique identifiers (effectively similar to a DOI) for research resources. This journal encourages authors to adopt RRIDs when reporting key biological resources (antibodies, cell lines, model organisms and tools) in their manuscripts.

Examples:

Organism: *Filip1*$^{tm1a(KOMP)Wtsi}$ **RRID:MMRRC_055641-UCD**

Cell Line: RST307 cell line **RRID:CVCL_C321**

Antibody: Luciferase antibody DSHB Cat# LUC-3, **RRID:AB_2722109**

Plasmid: mRuby3 plasmid **RRID:Addgene_104005**

Software: ImageJ Version 1.2.4 **RRID:SCR_003070**

RRIDs are provided by the Resource Identification Portal. Many commonly used research resources already have designated RRIDs. The portal also provides authors links so that they can quickly register a new resource and obtain an RRID.

Clinical Trial Registration

The World Health Organization (WHO) definition of a clinical trial is "any research study that prospectively assigns human participants or groups of humans to one or more health-related interventions to evaluate the effects on health outcomes". The WHO defines health interventions as “A health intervention is an act performed for, with or on behalf of a person or population whose purpose is to assess, improve, maintain, promote or modify health, functioning or health conditions” and a health-related outcome is generally defined as a change in the health of a person or population as a result of an intervention.

To ensure the integrity of the reporting of patient-centered trials, authors must register prospective clinical trials (phase II to IV trials) in suitable publicly available repositories. For example www.clinicaltrials.gov or any of the primary registries that participate in the WHO International Clinical Trials Registry Platform.

The trial registration number (TRN) and date of registration should be included as the last line of the manuscript abstract.

For clinical trials that have not been registered prospectively, authors are encouraged to register retrospectively to ensure the complete publication of all results. The trial registration number (TRN), date of registration and the words 'retrospectively registered’ should be included as the last line of the manuscript abstract.

Standards of reporting

Springer Nature advocates complete and transparent reporting of biomedical and biological research and research with biological applications. Authors are recommended to adhere to the minimum reporting guidelines hosted by the EQUATOR Network when preparing their manuscript.

Exact requirements may vary depending on the journal; please refer to the journal's Instructions for Authors.

Checklists are available for a number of study designs, including:

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Observational studies (STROBE)

Systematic reviews and meta-analyses (PRISMA) and protocols (Prisma-P)

Diagnostic/prognostic studies (STARD) and (TRIPOD)

Case reports (CARE)

Clinical practice guidelines (AGREE) and (RIGHT)

Qualitative research (SRQR) and (COREQ)

Animal pre-clinical studies (ARRIVE)

Quality improvement studies (SQUIRE)

Economic evaluations (CHEERS)

Summary of requirements

The above should be summarized in a statement and placed in a 'Declarations' section before the reference list under a heading of 'Ethics approval'.

Examples of statements to be used when ethics approval has been obtained:

• All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki Declaration and its later amendments or comparable ethical standards. The study was approved by the Bioethics Committee of the Medical University of A (No. ...).

• This study was performed in line with the principles of the Declaration of Helsinki. Approval was granted by the Ethics Committee of University B (Date.../No. ...).

• Approval was obtained from the ethics committee of University C. The procedures used in this study adhere to the tenets of the Declaration of Helsinki.

• The questionnaire and methodology for this study was approved by the Human Research Ethics committee of the University of D (Ethics approval number: ...).

Examples of statements to be used for a retrospective study:

• Ethical approval was waived by the local Ethics Committee of University A in view of the retrospective nature of the study and all the procedures being performed were part of the routine care.

• This research study was conducted retrospectively from data obtained for clinical purposes. We consulted extensively with the IRB of XYZ who determined that our study did not need ethical approval. An IRB official waiver of ethical approval was granted from the IRB of XYZ.

• This retrospective chart review study involving human participants was in accordance with the ethical standards of the institutional and national research committee and with the 1964 Helsinki Declaration and its later amendments or comparable ethical standards. The Human Investigation Committee (IRB) of University B approved this study.

Examples of statements to be used when no ethical approval is required/exemption granted:

• This is an observational study. The XYZ Research Ethics Committee has confirmed that no ethical approval is required.

• The data reproduced from Article X utilized human tissue that was procured via our Biobank AB, which provides de-identified samples. This study was reviewed and deemed exempt by our XYZ Institutional Review Board. The BioBank protocols are in accordance with the ethical standards of our institution and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

Authors are responsible for correctness of the statements provided in the manuscript. See also Authorship Principles. The Editor-in-Chief reserves the right to reject submissions that do not meet the guidelines described in this section.

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Informed consent

All individuals have individual rights that are not to be infringed. Individual participants in studies have, for example, the right to decide what happens to the (identifiable) personal data gathered, to what they have said during a study or an interview, as well as to any photograph that was taken. This is especially true concerning images of vulnerable people (e.g. minors, patients, refugees, etc) or the use of images in sensitive contexts. In many instances authors will need to secure written consent before including images.

Identifying details (names, dates of birth, identity numbers, biometrical characteristics (such as facial features, fingerprint, writing style, voice pattern, DNA or other distinguishing characteristic) and other information) of the participants that were studied should not be published in written descriptions, photographs, and genetic profiles unless the information is essential for scholarly purposes and the participant (or parent/guardian if the participant is a minor or incapable or legal representative) gave written informed consent for publication. Complete anonymity is difficult to achieve in some cases. Detailed descriptions of individual participants, whether of their whole bodies or of body sections, may lead to disclosure of their identity. Under certain circumstances consent is not required as long as information is anonymized and the submission does not include images that may identify the person.

Informed consent for publication should be obtained if there is any doubt. For example, masking the eye region in photographs of participants is inadequate protection of anonymity. If identifying characteristics are altered to protect anonymity, such as in genetic profiles, authors should provide assurance that alterations do not distort meaning.

Exceptions where it is not necessary to obtain consent:

• Images such as x rays, laparoscopic images, ultrasound images, brain scans, pathology slides unless there is a concern about identifying information in which case, authors should ensure that consent is obtained.

• Reuse of images: If images are being reused from prior publications, the Publisher will assume that the prior publication obtained the relevant information regarding consent. Authors should provide the appropriate attribution for republished images.

Consent and already available data and/or biologic material

Regardless of whether material is collected from living or dead patients, they (family or guardian if the deceased has not made a pre-mortem decision) must have given prior written consent. The aspect of confidentiality as well as any wishes from the deceased should be respected.

Data protection, confidentiality and privacy

When biological material is donated for or data is generated as part of a research project authors should ensure, as part of the informed consent procedure, that the participants are made aware what kind of (personal) data will be processed, how it will be used and for what purpose. In case of data acquired via a biobank/biorepository, it is possible they apply a broad consent which allows research participants to consent to a broad range of uses of their data and samples which is regarded by research ethics committees as specific enough to be considered "informed". However, authors should always check the specific biobank/biorepository policies or any other type of data provider policies (in case of non-bio research) to be sure that this is the case.

Consent to Participate

For all research involving human subjects, freely-given, informed consent to participate in the study must be obtained from participants (or their parent or legal guardian in the case of children under 16) and a statement to this effect should appear in the manuscript. In the case of articles describing human transplantation studies, authors must include a statement declaring that no organs/tissues were obtained from prisoners and must also name the institution(s)/clinic(s)/department(s) via which organs/tissues were obtained. For manuscripts reporting studies involving vulnerable groups where there is the potential for coercion or where consent may not have been fully informed, extra care will be taken by the editor and may be referred to the Springer Nature Research Integrity Group.

Consent to Publish

Individuals may consent to participate in a study, but object to having their data published in a journal article. Authors should make sure to also seek consent from individuals to publish their data prior to submitting their paper to a journal. This is in particular applicable to case studies. A consent to publish form can be found

here. (Download docx, 36 kB)

Summary of requirements

The above should be summarized in a statement and placed in a 'Declarations' section before the reference list under a heading of 'Consent to participate' and/or 'Consent to publish'. Other declarations include Funding, Competing interests, Ethics approval, Consent, Data and/or Code availability and Authors' contribution statements.

Please see the various examples of wording below and revise/customize the sample statements according to your own needs.

Sample statements for **"Consent to participate"**:

Informed consent was obtained from all individual participants included in the study.

Informed consent was obtained from legal guardians.

Written informed consent was obtained from the parents.

Verbal informed consent was obtained prior to the interview.

Sample statements for **"Consent to publish"**:

The authors affirm that human research participants provided informed consent for publication of the images in Figure(s) 1a, 1b and 1c.

The participant has consented to the submission of the case report to the journal.

Patients signed informed consent regarding publishing their data and photographs.

Sample statements if identifying information about participants is available in the article:

Additional informed consent was obtained from all individual participants for whom identifying information is included in this article.

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Images will be removed from publication if authors have not obtained informed consent or the paper may be removed and replaced with a notice explaining the reason for removal.

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Research involving animals, their data or biological material

The welfare of animals (vertebrate and higher invertebrate) used for research, education and testing must be respected. Authors should supply detailed information on the ethical treatment of their animals in their submission. For that purpose they may use the ARRIVE checklist which is designed to be used when submitting manuscripts describing animal research.

For studies involving client-owned animals, authors must also document informed consent from the client or owner and adherence to a high standard (best practice) of veterinary care.

Authors are recommended to comply with:

• The International Union for Conservation of Nature (IUCN) Policy Statement on Research Involving Species at Risk of Extinction and consult the IUCN red list index of threatened species.

• Convention on the Trade in Endangered Species of Wild Fauna and Flora

When reporting results authors should indicate:

• … that the studies have been approved by a research ethics committee at the institution or practice at which the studies were conducted. Please provide the name of ethics committee and relevant permit number;

• … whether the legal requirements or guidelines in the country and/or state or province for the care and use of animals have been followed.

Researchers from countries without any legal requirements or guidelines voluntarily should refer to the following sites for guidance:

– The Basel Declaration describes fundamental principles of using animals in biomedical research

– The International Council for Laboratory Animal Science (ICLAS) provides ethical guidelines for researchers as well as editors and reviewers

– The Association for the study of Animal Behaviour describes ethical guidelines for the treatment of animals in research and teaching

– The International Association of Veterinary Editors' Consensus Author Guidelines on Animal Ethics provide guidelines for authors on animal ethics and welfare

Researchers may wish to consult the most recent (ethical) guidelines available from relevant taxon-oriented professional societies.

If a study was granted exemption or did not require ethics approval, this should also be detailed in the manuscript.

Summary of requirements

The above should be summarized in a statement and placed in a 'Declarations' section before the reference list under a heading of 'Ethics approval'.

Please see the various examples of wording below and revise/customize the sample statements according to your own needs.

Examples of statements to be used when ethics approval has been obtained:

• All procedures involving animals were in compliance with the European Community Council Directive of 24 November 1986, and ethical approval was granted by the Kocaeli University Ethics Committee (No. 29 12 2014, Kocaeli, Turkey).

• All procedures performed in the study were in accordance with the ARVO Statement for Use of Animals in Ophthalmic Vision and Research. The ethical principles established by the National Institutes of Health Guide for the Care and Use of Laboratory Animals (NIH Publications No. 8523, revised 2011) were followed. The research protocol was approved by the Ethics Committee on Animal Use (Protocol No. 06174/14) of FCAV/Unesp, Jaboticabal.

• This study involved a questionnaire-based survey of farmers as well as blood sampling from their animals. The study protocol was assessed and approved by Haramaya University, research and extension office. Participants provided their verbal informed consent for animal blood sampling as well as for the related survey questions. Collection

of blood samples was carried out by veterinarians adhering to the regulations and guidelines on animal husbandry and welfare.

• All brown bear captures and handling were approved by the Ethical Committee on Animal Experiments, Uppsala, Sweden (Application C18/15) and the Swedish Environmental Protection Agency in compliance with Swedish laws and regulations.

• The ethics governing the use and conduct of experiments on animals were strictly observed, and the experimental protocol was approved by the University of Maiduguri Senate committee on Medical Research ethics. Proper permit and consent were obtained from the Maiduguri abattoir management, before the faecal samples of the cattle and camels slaughtered in this abattoir were used for this experiment.

Examples of statements to be used when no ethical approval is required/exemption granted:

• No approval of research ethics committees was required to accomplish the goals of this study because experimental work was conducted with an unregulated invertebrate species.

• As the trappings of small mammals were conducted as part of regular pest control measures in accordance with the NATO Standardized Agreement 2048 "Deployment Pest and Vector Surveillance and Control ", no approval by an ethics committee was required.

• All experiments have been conducted as per the guidelines of the Institutional Animal Ethics Committee, Department of Zoology, Utkal University, Bhubaneswar, Odisha, India. However, the insect species used in this study is reared for commercial production of raw silk materials, as a part of agro-based industry. Therefore, use of this animal in research does not require ethical clearance. We have obtained permission from the office of Research officer sericulture, Baripada, Orissa, India for the provision of infrastructure and support for rearing of silkworm both in indoor and outdoor conditions related to our study to promote sericulture practices.

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Funding and Support pages

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Research Data Policy and Data Availability Statements

This journal operates a type 3 research data policy. A submission to the journal implies that materials described in the manuscript, including all relevant raw data, will be freely available to any researcher wishing to use them for non-commercial purposes, without breaching participant confidentiality.

Data availability

All original research must include a data availability statement. Data availability statements should include information on where data supporting the results reported in the article can be found, if applicable. Statements should include, where applicable, hyperlinks to publicly archived datasets analysed or generated during the study. For the purposes of the data availability statement, "data" is defined as the minimal dataset that would be necessary to interpret, replicate and build upon the findings reported in the article. When it is not possible to share research data publicly, for instance when individual privacy could be compromised, data availability should still be stated in the manuscript along with any conditions for access. Data availability statements can take one of the following forms (or a combination of more than one if required for multiple datasets):

1. The datasets generated during and/or analysed during the current study are available in the [NAME] repository, [PERSISTENT WEB LINK TO DATASETS]

2. The datasets generated during and/or analysed during the current study are available from the corresponding author on reasonable request.

3. All data generated or analysed during this study are included in this published article [and its supplementary information files].

4. The datasets generated during and/or analysed during the current study are not publicly available due [REASON(S) WHY DATA ARE NOT PUBLIC] but are available from the corresponding author on reasonable request.].

5. Data sharing not applicable to this article as no datasets were generated or analysed during the current study.

6. The data that support the findings of this study are available from [THIRD PARTY NAME] but restrictions apply to the availability of these data, which were used under licence for the current study, and so are not publicly available. Data are however

available from the authors upon reasonable request and with permission of [THIRD PARTY NAME].

More templates for data availability statements, including examples of openly available and restricted access datasets, are available here:

Data availability statements

Data repositories

This journal strongly encourages that all datasets on which the conclusions of the paper rely are available to readers. We encourage authors to ensure that their datasets are either deposited in publicly available repositories (where available and appropriate) or presented in the main manuscript or additional supporting files whenever possible. Please see Springer Nature's information on recommended repositories.

List of Repositories

General repositories - for all types of research data - such as figshare and Dryad may be used where appropriate.

Data citation

The journal also requires that authors cite any publicly available data on which the conclusions of the paper rely. Data citations should include a persistent identifier (such as a DOI), should be included in the reference list using the minimum information recommended by DataCite, and follow journal style. Dataset identifiers including DOIs should be expressed as full URLs.

Research data and peer review

Peer reviewers are encouraged to check the manuscript's Data availability statement, where applicable. They should consider if the authors have complied with the journal's policy on the availability of research data, and whether reasonable effort has been made to make the data that support the findings of the study available for replication or reuse by other researchers. Peer reviewers are entitled to request access to underlying data (and code) when needed for them to perform their evaluation of a manuscript.

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authorship in the data is also blinded. There are data repositories that can assist with this and/or will create a link to mask the authorship of your data.

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