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Thermotolerant Capacity of Diverse Indonesian Lowland Holstein's Black and White Skin-Pigmentation Intensity

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ABSTRACT

The lowland area of Indonesia has economic potential to develop as a major centre for dairy farming businesses. Still, there is a foremost matter: overheated irradiation immersed in damped air would increase the heat stress level. The present study was conducted to assess the heat stress tolerance level of lowland Indonesian Holstein cows, characterized by a dominant level of Black and White (B/W) pigmented skin coat. This study involved 34 heads of Holstein cows that were cared for in Eastern Jakarta. Breathing frequency (BF), rectum's warmth (RW), pulse rate (PL), white-skinned spot temperature (WSST), black-skinned spot temperature (BSST), average temperature rounded skin (ATRS), back region temperature (BRT), chest region temperature (CRT), upper-leg temperature (ULT), lower-leg temperature (LLT), skin's temperature (ST), body's temperature (BT), and heat tolerance coefficient (HTC) as variables were checked statistically with R 4.4.2 type software comprised correlation and independent t-test. Cows B/W classification performed with the imageJ 1.54g series program. Those analyses showed that most lowland Indonesian Holsteins were characterized by black-pigmented dominant cows (BDC). The pigmentation of both lateral sides of the body generated a significant correlation ($p<0.05$ and $p<0.01$) with several vital symptoms, including WSST, ATRS, ST, BT, and HTC, but only in the BDC Holstein class. The test of mean distinction between the two classes (BDC and WDC) produced statistically insignificant differences ($p>0.05$) in all measurable vital symptoms; all variables showed trends indicating that the BDC class had a greater response to heat stress. The BDC Holstein type selection base had high expectations of elevating the HTC score in Indonesian lowland cows, as the closing decision.

Keywords: *Coastline region, Dairy cattle, Heat stress, Humid, Tropical*

Introduction

An archipelago country like Indonesia has an extensive coastline around the islands (Djunarsjah and Putra, 2021). In addition, Indonesia is located in the equatorial line region; consequently, it has merely two annual seasons: dry and rainy (Kurniawan *et al*., 2024). Thus, the microclimate and macroclimate in the coastal area during wet tropical weather are likely to cause heatstroke on a daily basis and throughout the year (Setiawati *et al*., 2022). A case in Jakarta in 2019 had a temperature minimum (T_{min}), maximum (T_{max}), average (T_{ave}), and relative humidity (rH) of 29.25ºC, 40.00ºC, 36.39ºC, and 78% (Wijayanto *et al*., 2020).

Inevitably, the majority of Indonesian citizens' residences are located in lowland areas and close to the coastline (Putri *et al*., 2018). A scholarly paper has proven that Indonesia ranks fifth among countries with the highest subsistence rate in the lowland area worldwide, affecting approximately 42% of the total population (Neumann *et al*., 2015). These phenomena would shift the amount of dietary consumption in lowland urban areas, including dairy products (Nurhasan *et* *al*., 2022). Still, over 98% of the Indonesian Holstein cattle population is currently nurtured on Java Island (Asmarasari *et al*., 2023) and is prevalent in the highland region, which has comparative advantages in ambient temperature over the lowland (Iskandar, 2011). However, the available square area is limited. Again, the thermal humidity index (THI) of barn microclimates and the heat tolerance coefficient (HTC) of cattle have an impact on milk production (Rachmawati *et al*., 2024). A report recommends relocating the dairy industry to another island, such as Sumatra, to achieve sustainable development goals (SDGs) in the dairy sector of Indonesia (Apdini *et al*., 2024). Countries like Indonesia, which mirror climate conditions, propose intervening in cattle diets and genetic aspects as the focal point to address milk yield gaps (Mayberry *et al*., 2017). Reasonably, the Indonesian Holstein should be genetically engineered for improved performance in thermotolerance to better cope with heat stress challenges (Tessalonika *et al*., 2024).

Performing a selection in the Indonesian Holstein linked with HTC could enhance thermotolerant capacity. For instance, black and white skin pigmentation in Holstein is allegedly associated with several production traits like longevity (Baek and Lee, 2015; Lee *et al*., 2016), potential adaptation aptitude (Laible *et al*., 2021), evolution mechanism (Eizirik and Trindade, 2021), physiological status (Putra *et al*., 2021), milk delivery (Maia, da Silva, Bertipaglia, *et al*., 2005), body gain and development (Rashid *et al*., 2019), and amount of thermal radiative transmittance, reflection, and absorption (Maia, da Silva, and Bertipaglia, 2005). This experimental design offers skin coat pigmentation intensity selection based on its affordability, ease of handling, simplicity, and lack of specialized skills, yet it is potent and reliable. Despite evidence linking coat colour with heat tolerance, little is known about this relationship in Indonesian Holsteins. Data disclosure regarding the impression of skin coat colour position and heat tolerance capacity (HTC) is urgently needed to initiate a breeding program for an exclusive cow for a lowland tropical region, which serves as the ultimate goal of this exploration.

Materials and Methods

Samples and variables profile

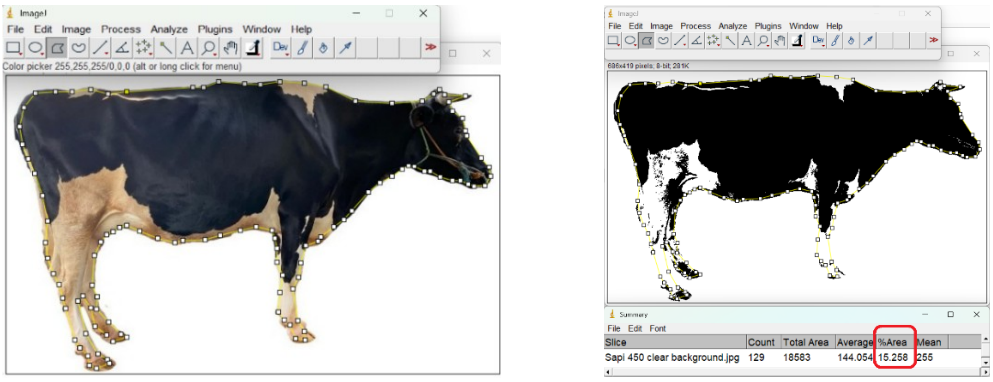
Exploration was conducted in the administrative city of East Jakarta, specifically at Pondok Ranggon Village, Cipayung subdistrict. This coordinate was approximately -2 to +50 meters above sea level (MSL) (Luo *et al*., 2019). The investigation involved a sample of 34 Indonesian Holstein cows from a smallholder dairy farm (SDF). Cows' ages ranged from 2 to 6 years in the lactation period and were fed with similar feed ingredients, which included baby corn forages, commercial concentrate, and okara. Chiefly, this study was divided into two primary data sets encompassing information on cattle B/W pigmentation levels and their physiological responses. All data assemblage procedures were undertaken during this exploration, which was already permitted by the IACUCs of SVMBS-IPB University, as documented in document no. 283/KEH/SKE/I/2025. The breakdown variables are presented in the following paragraph.

The checked variable was the black-white (B/W) intensity of Indonesian Holstein using five frames of reference, consisting of left lateral view (LLV), right lateral view (RLV), upper view (UPV), hind view (HNV), and body rounded view (BRV), and the investigators captured it with a camera to generate a digital image. Settled at a resolution of 96 dpi and 4032 x 2268 pixels, the image of the cows was captured after the body was washed to remove stuck materials from the skin coat, thereby minimizing bias. The recognition frame of the dairy cow is often investigated using individual digital photographs, and it has been reliably proven (Andrew *et al*., 2021; Kim *et al*., 2005; Nasution *et al*., 2024). Antecedent variables were characterized as independent factors. Meanwhile, the dependent constituents include breathing frequency (BF), rectum's warmth (RW), pulses (PL), White-skinned spot temperature (WSST), black-skinned spot temperature (BSST), Average temperature rounded skin (ATRS), back region temperature (BRT), chest region temperature (CRT), upper-leg temperature (ULT), lower-leg temperature (LLT), skin's temperature (ST), body's temperature (BT), and heat tolerance coefficient (HTC). Afterwards, dependent and independent elements were distributed in a dichotomous column according to the preponderance of B/W colour of skin pigmentation intensity, respectively. Additional information, such as the barn microclimate temperature (Ta) and relative humidity (rH), was captured by inspectors using a digital thermo-humidity data logger set to an interval of 15 minutes. The logger was positioned in the middle area of the cowshed, 2 meters from the concrete, to determine the ambient conditions. Subsequently, the degree of the thermo-humidity index (THI) was calculated to assess the level of heatstroke exposure in cows.

B/W pigmentation level segregation method

The inspector analyzed the cow's digital image using public domain software called ImageJ 1.54g series to determine the dominance intensity of skin-coat colour (black and white) for each. In advance, the raw digital image of the cows was cleaned to remove the disturbing background. In this program, the area of the raw digital image to be quantified was selected and subsequently converted into an 8-bit image type, after which an auto-threshold was applied. Finally, the analysis particles procedure was run. The first figure illustrates the cow's digital image processing with ImageJ software and the outcome of reading etiquette.

Fig. 1(A) and 1(B) are instances of the B/W skin-pigmentation quantification procedure outlined in the percentage score. Recorded in Fig. 1(B), send proof that this cow's digital image had as much as 15.26% white skin pigmentation, and the remainder, 84.74%, is black. This procedure was performed from the four points of view of each cow's digital images, adjunct to the average accumulation score of those four sceneries as a fifth perspective. Subsequently, discrimination was performed to classify the sample based on the predominant level of B/W coat pigmentation, where less than or equal to 50% white color is categorized as a black-dominant's cow (BDC), and greater than 50% white is a white-dominant's cow (WDC); this approach prevails in a reciprocal mechanism and is held by one of the category setups following the Lee method (Lee *et al.*, 2016).



(A) (B)

Fig. 1. The B/W-dominated-skinned percentage pigmentation segregation method of cows (A) cleanses the background image of the cow, and (B) the cow picture product after testing.

Variables math formulas

Since the sample cattle had been identified with the proportion of B/W colouring individually, the BF, RW, PL, WSST, BSST, AST, BRT, CRT, ULT, LLT, ST, BT, and HTC variables were input accordingly with those categories. Circumstantially, the RW measurement utilized a digital thermometer with an accuracy of ±0.1ºC in the range of 32.0 to 42.0ºC, PL quantification with a stethoscope, and BF was calculated manually based on the ex-inhale frequency. Meanwhile, WSST, BSST, ATRS, BRT, CRT, ULT, and LLT were sized using a digital infrared thermometer with an accuracy of ±0.1ºC in the range of 32.0 to 42.0ºC. Remnants, the ST, BT, HTC, and THI values were estimated to follow the mathematical models (a), (b), (c), and (d) seriatim. Chiefly for BT and ST computerized with McLean's method:

(a) (McLean *et al.*, 1983)

$$\mathrm{ST} = 0.25\,(\mathrm{BRT} + \mathrm{CRT}) + 0.32\,\mathrm{ULT} + 0.18\,\mathrm{LLT}$$

ST : skin's temperature (ºC);
BRT : back region's temperature (ºC);
CRT : chest region's temperature (ºC);
ULT : upper-leg's temperature (ºC); and
LLT : lower-leg's temperature (ºC).

(b) (McLean *et al.*, 1983)

$$\mathrm{BT} = 0.86\,\mathrm{RW} + 0.14\,\mathrm{ST}$$

BT : body's temperature (ºC);
RW : rectum's warmth (ºC);
ST : skin's temperature (ºC).

Meanwhile, the HTC calculation follows the established model by Rhoads:

(c) (Gaalaas, 1947)

$$\mathrm{HTC} = 100 - [14(\mathrm{BT} - 101)]$$

HTC : heat tolerance coefficient; and
BT : body temperature (ºF).

Since the BT computation is generated in Celsius degrees, it should be converted to Fahrenheit degrees in advance. A score greater than 100 points on the HTC signified better thermotolerance aptness; otherwise, a lower score adversely affected it. Next, the mathematical model to take into account THI follows this model:

(d) (Yeck *et al.*, 1971)

$$\mathrm{THI} = (1.8 \times \mathrm{Ta} + 32) - (0.55 - 0.0055 \times \mathrm{rH}) \times (1.8 \times \mathrm{Ta} - 26.6)$$

THI : thermo-humidity index;
T_a :ambient temperature (ºC); and
rH : relative humidity (%).

In the condition score, a THI below 72 indicates that the cows are unexposed to heat stress, 72–79 means they are exposed to mild heat stress, 80–89 means they are exposed to moderate heat stress, and a value greater than 90 indicates they are exposed to severe heat stress.

Statistical scrutiny approach

Under the data status, the data were intact, accumulated, well-organized, and consistent with the BDC and WDC allocations. A Shapiro-Wilk normality test was executed in advance to identify the data distribution. They were then followed by statistical analysis, including descriptive statistics, correlation, and independent t-tests. This statistical scrutiny was tested using R 4.4.2, an open-source software, and RStudio.

Statistical descriptive minimum, mean, and maximum scores were used to scrutinize the cowshed microclimates' ambient data, physiological symptoms rate, and skin coat pigmentation distribution. The correlation test is used to quantify the level of relationship among the four body perspectives and within their physiological rates. A mean difference test was conducted to compare physiological symptoms between the BDC and WDC classes.

Results and Discussion

Ambient barn microclimate exposure

Determining the cow's ambient heat exposure level is an excellent starting point for this exploration. Micro- and macroclimate temperature and humidity exposure levels transmit heat to the cows; subsequently, it leverages the stress scale undergone (West, 2003). Dairy animals experience reduced feed intake and milk production, poor reproductive function, and an increased risk of health concerns during hot weather, all of which impact output and their overall well-being (Chen *et al*., 2025). A detailed description of the barn's microclimate, or the area's internal environment, encompassing temperature, relative humidity, thermal humidity index, and stress exposure level, is provided in Table 1.

Table 1. Daytime, nighttime, and full-day Indonesia lowland barn microclimate conditions

Period	Ta (ºC)	rH (%)	THI	Stress level
Daytime (12-h)	32.03 ± 1.57	62.63 ± 8.15	83.01 ± 1.24	Moderate
Nighttime (12-h)	26.33 ± 1.11	85.27 ± 5.27	77.63 ± 1.60	Mild
Fullday (24-h)	29.12 ± 1.05	74.57 ± 5.71	80.68 ± 1.18	Moderate

Ta: temperature; rH: relative humidity; THI: thermal humidity index; 12-h: 12 hours; and 24-h: 24 hours.

Microclimate metering results showed that Indonesian cows in the lowland area were exposed to distinct heat stroke levels during both day and night. A superior catastrophe effect was observed during the daytime compared to nighttime, despite only a one-level difference. This situation occurred because the daytime temperature was higher than the nighttime temperature, but it had a more comfortable relative humidity level. Between daytime and nighttime, there was an interchange of those phenomena. The heatstroke alert was also intensely expressed in the data on vital symptoms published in Table 2, particularly in the BF and PL rates. They were overscored compared with the standard range frequency, which a cow should pound. An Indonesian researcher found that the normal range of cow BF and PL is within 26-37 times per minute and 59-72 times per minute, respectively (Suprayogi *et al*., 2017). The measurable RW, ST, and BT warmth were within the normal range that a cow should emit. While the HTC score was generally greater than 100 points, only two individuals scored lower. WDC Holstein, fascinatingly, possessed the lowest level of HTC points. Thus, diverse barn ambient management between daytime and nighttime has a strong possibility of helping to shift the exposure level of heatstroke in cows. For instance, the daytime could be focused on the barn temperature management system, while concentrating on the barn relative humidity management system at night. Indeed, combined with better thermotolerant dexterity, it would significantly increase the cow's comfort level. Table 3 presents the sample distribution of B/W-skinned-dominated level cows to determine the HTC level.

Table 2. Indonesian lowland Holstein cows' physiological response rate of the total samples

Vital symptoms	Minimum	Mean±SD	Maximum
BF (times minutes^{-1})	50.67	65.60 ± 9.32	86.68
PL (times minutes^{-1})	67.56	86.95 ± 8.83	106.20
RW (ºCelcius)	37.78	38.39 ± 0.38	39.40
ST (ºCelcius)	32.55	33.36 ± 0.35	33.97
BT (ºCelcius)	37.12	37.69 ± 0.35	38.62
HTC	92.67	116.29 ± 8.70	130.56

BF: breathing frequency; PL: pulse rate; RW: rectum's warmth; ST: skin's temperature; BT: body's temperature; HTC: heat tolerance coefficient; and SD: standard deviation.

Table 3. Cow's sample distribution according to the domination level of B/W-skinned pigmentation in lowland Indonesia

Viewpoint classification	BDC (Mean±SE)	WDC (Mean±SE)
White left lateral view (WLLV) (%)	22.17 ± 2.45	71.44 ± 7.72
Black left lateral view (BRLV) (%)	77.83 ± 2.45	28.56 ± 7.72
White right lateral view (WRLV) (%)	23.76 ± 2.61	71.80 ± 6.20
Black right lateral view (BRLV) (%)	76.24 ± 2.61	28.20 ± 6.20
White upper view (WUPV) (%)	22.43 ± 3.51	70.83 ± 11.65
Black upper view (BUPV) (%)	77.57 ± 3.51	29.17 ± 11.65
White hind view (WHNV) (%)	18.83 ± 3.02	78.82 ± 4.39
Black hind view (BHNV) (%)	81.17 ± 3.02	21.18 ± 4.39
White body rounded view (WBRV) (%)	21.80 ± 2.37	73.22 ± 5.48
Black body rounded view (BBRV) (%)	78.20 ± 2.37	26.78 ± 5.48

BDC: black-skinned dominated cows; WDC: white-skinned dominated cows; and SE: standard error.

Ambient temperature exceeding 35ºC promptly triggered the cattle thermoregulation switch-button (Berman, 2005). Although the daily temperature of the current exploration was still below that score, the Indonesian lowland Holstein cows were exposed to moderate heat stress due to the moist microclimate. An effective response to heat stress issues is a focusable selection method based on physiological features (Cartwright *et al*., 2023). Cattle under heat stress in an ambient environment would increase their breathing frequency (BF) and rectal temperature (RW) to 44-89 times per minute and 38.6-40.4ºC, respectively (Wheelock *et al*., 2010). Breathing frequency is a detectable indicator of heatstroke responses (Berman, 2006). While the cow's body thermo-neutral regulation system constantly struggles to maintain the RW within the 38-39.2ºC range (Ammer *et al*., 2016), surpassing 39.3ºC is a vigilant condition (Pragna *et al*., 2017). The BF above 60 times per minute is a poor signal for the cow's thermoregulation regarding heatstroke (Das *et al*., 2016). Since the BF and RW escalated beyond the standard, the pulse rate (PL) is gradually promoted as a response to the shift in the number of respiratory muscle contractions and the blood supply mechanism to the dermis region to initiate the heat transfer process intensely, with the consequence of vasodilation (Yameogo *et al*., 2021), interval standard of cows heart rate is 54-84 pulsation per minute (Chen *et al*., 2022).

Expression of B/W pigmentation class

After skin-pigmentation analysis and classification were performed, it was found that 27 of the 34 cows were dominated by black-skinned pigmentation, and seven cows were dominated by white-skinned pigmentation. Impartially, the Indonesian lowland area was ruled by the BDC Holstein. Interestingly, the facts also indicate, based on the data in Table 3, that the percentage of inequality scores among the four viewpoints varied. Despite the left and right lateral sides, a comparison was made in both tabulations. Appealing to the BDC and WDC Holstein groups, they presented data showing that the BDC was superior on both lateral sides, as well as in upper and hind views. The rounded perspectives' average score indicated a similar tendency. This evidence strengthened the claims that Holstein cattle with BDC traits were prominent in the sample population of the lowlands of Indonesia. A breakdown analysis of the correlation level between various viewpoints and percentages of the underlying B/W-dominated skinned point is presented in Table 4 to disclose which body area positions have connections and interdependencies. Subsequently, a deeper scrutiny was conducted between the four viewpoints of the percentage of skinned pigmentation on cattle bodies and the measurable vital symptom scores in Table 5 to assess the statistical interconnection level.

Table 4. Correlation degree between the four viewpoints on the pigmentation of the B/W-skinned-dominated Indonesian Holsteins group in the lowland

Viewpoint	WRLV	BRLV	WLLV	BLLV	WUPV	BUPV	WHNV	BHNV	WBRV	BBRV
WRLV	1.00	-1.00**	-0.95**	0.95**	0.56**	-0.56**	0.36	-0.36	0.85**	-0.85**
BRLV	-1.00**	1.00	0.95**	-0.95**	-0.56**	0.56**	-0.36	0.36	-0.85**	0.85**
WLLV	0.83*	-0.83*	1.00	-1.00**	0.60**	-0.60**	0.45*	-0.45*	0.89**	-0.89**
BLLV	-0.83*	0.83*	-1.00**	1.00	-0.60**	0.60**	-0.45*	0.45*	-0.89**	0.89**
WUPV	0.78*	-0.78*	0.85*	-0.85*	1.00	-1.00**	0.49*	-0.49*	0.84**	-0.84**
BUPV	-0.78*	0.78*	-0.85*	0.85*	-1.00**	1.00	-0.49*	0.49*	-0.84**	0.84**
WHNV	-0.62	0.62	-0.49	0.49	0.55	-0.55	1.00	-1.00**	0.72**	-0.72**
BHNV	0.62	-0.62	0.49	-0.49	-0.55	0.55	-1.00**	1.00	-0.72**	0.72**
WBRV	0.87*	-0.87*	0.95**	0.95**	0.94**	-0.94**	-0.44	0.44	1.00	-1.00**
BBRV	-0.87*	0.87*	-0.95**	0.95**	-0.94**	0.94**	0.44	-0.44	-1.00**	1.00

The up-zone of the slanted cross belongs to black-skinned-dominated cows; the downward zone belongs to white-skinned-dominated cows. **: significant on p<0.01; *: significant on p<0.05; WRLV: white right lateral viewpoint; BRLV: black right lateral viewpoint; WLLV: white left lateral viewpoint; BLLV: black left lateral viewpoint; WUPV: white upper viewpoint; BUPV: black upper viewpoint; WHNV: white hind viewpoint; BHNV: black hind viewpoint; WBRV: white body rounded viewpoint; BBRV: black body rounded viewpoint.

A statistical program's result revealed that the highest mark on the BDC Holstein group was found in the relationship between the pigmentation percentage of the B/W-skinned rooted within the left (WLLV and BLLV) and right (WRLV and BRLV) lateral sides. In contrast, the lowest values were observed in the hind area (WHNV and BHNV) and on the right lateral side (WRLV and BRLV). On the WDC Holstein cluster, the sublime connection was observed on the left lateral sides (WLLC and BLLV) in conjunction with the upper side (WUPV and BUPV), in contrast to the shortest hind side (WHNV and BHNV) and the left lateral side (WLLC and BLLV). A parallel phenomenon stood out in the rounded average score of B/W-skinned pigmentation percentage, a zenithal connection between the rounded view (WBRV and BBRV) and the left lateral sides (WLLV and BLLV). The rounded view (WBRV and BBRV) achieved the highest bottom link score in the hind area (WHNV and BHNV). Those entire data calls were explicitly exposed in Table 4.

The origin of variations in skin tone among individuals worldwide is attributed to pigmentation on the dermis surface and coat (Bastonini *et al*., 2016). Skin-coat pigmentation results from melanocytes, which give the skin its colour and shield it from detrimental UV rays (UVR) damage (Lin and Fisher, 2007; Serre *et al*., 2018). They are also accountable for colouring (Yamaguchi *et al*., 2007). The melanogenesis process predisposed the opacity level of the skin-coat pigment to produce melanin (Videira *et al*., 2013). Melanin is essential for shielding genes, the epidermis and tissues against UV-induced skin harm (Hamblin, 2023). Melanin is well-known for its role as "photoprotection" from UVR to minimize the formation of cancer cells on the skin (Hearing, 2011). Melanocytes are likely involved in a wide range of physiological processes, and their specific roles in each target location go much beyond the synthesis of melanin alone (Ali and Naaz, 2015). In some individuals, it serves as camouflage, thermo-regulation, and other functions (Hearing, 2011). In detail, the melanism scheme is committed to thermoregulation, intraspecific communication, defence against parasites, crypsis (defence against visual predators and camouflage facilitating ambush predation), and coding or regulatory polymorphisms in ASIP, MC1R, and CBD103 (Eizirik and Trindade, 2021). The sort and quantity of melanin pigment that melanocytes make and distribute to keratinocytes determines the tone of an individual's skin (Bastonini *et al*., 2016). The fibroblast contributes to that mechanism as an overlay (Hamblin, 2023; Widgerow *et al*., 2022).

Table 5. The correlation degree of four viewpoints on the pigmentation of the B/W-skinned dominated group with vital symptoms of Holsteins nursed in the lowland Indonesian area.

	Parameters	WRLV	BRLV	WLLV	BLLV	WUPV	BUPV	WHNV	BHNV	WBRV	BBRV
BDC	BF	0.22	-0.22	0.20	-0.20	0.30	-0.30	-0.05	0.05	0.21	-0.21
	PL	0.15	-0.15	0.11	-0.11	-0.01	0.01	-0.07	0.07	0.04	-0.04
	RW	0.34	-0.34	0.24	-0.24	0.10	-0.10	0.00	-0.00	0.19	-0.19
	WSST	0.43*	-0.43*	0.49**	-0.49**	0.32	-0.32	0.12	-0.12	0.40*	-0.40*
	BSST	0.33	-0.33	0.42*	-0.42*	0.22	-0.22	0.07	-0.07	0.30	-0.30
	ATRS	0.39*	-0.39*	0.47*	-0.47*	0.28	-0.28	0.10	-0.10	0.36	-0.36
	ST	0.44*	-0.44*	0.46*	-0.46*	0.09	-0.09	0.03	-0.03	0.28	-0.28
	BT	0.39*	-0.39*	0.30	-0.30	0.11	-0.11	0.01	-0.01	0.27	-0.27
	HTC	0.39*	-0.39*	-0.30	0.30	-0.11	0.11	-0.01	0.01	-0.22	0.22
WDC	BF	0.20	-0.20	0.21	-0.21	0.59	-0.59	-0.35	-0.35	0.37	-0.37
	PL	-0.13	0.13	-0.31	0.31	0.10	-0.10	-0.23	-0.23	-0.14	0.14
	RW	-0.05	0.05	-0.11	0.11	0.24	-0.24	-0.32	-0.32	0.01	-0.01
	WSST	-0.55	0.55	-0.36	0.36	-0.43	0.43	0.47	0.47	-0.42	0.42
	BSST	-0.45	0.45	-0.27	0.27	-0.39	0.39	0.52	0.52	-0.32	0.32
	ATRS	-0.50	0.50	-0.31	0.31	-0.42	0.42	0.49	0.49	-0.38	0.38
	ST	-0.12	0.12	0.20	-0.20	0.20	-0.20	0.02	0.02	0.15	-0.15
	BT	-0.05	0.05	-0.07	0.07	0.25	-0.25	-0.29	-0.29	0.03	-0.03
	HTC	0.06	-0.06	0.08	-0.08	-0.25	0.25	0.29	0.29	-0.03	0.03

WRLV: white correct lateral viewpoint; BRLV: black right lateral viewpoint; WLLV: white left lateral viewpoint; BLLV: black left lateral viewpoint; WUPV: white upper viewpoint; BUPV: black upper viewpoint; WHNV: white hind viewpoint; BHNV: black hind viewpoint; WBRV: white body rounded viewpoint; BBRV: black body rounded viewpoint; BDC: black-skinned dominated cows; WHC: white-skinned dominated cows; BF: breathing frequency; PL: pulse rate's; RW: rectum's warmth; WSST: white-skinned spot temperature; BSST: black-skinned spot temperature; ATRS: Average temperature rounded skin; ST: skin's temperature; BT: body's temperature; HTC: heat tolerance coefficient; **: significant on p<0.01; *: significant on p<0.05.

Relationship level of vital symptoms underlying the B/W class

Information concerning the relationship between the percentage of pigmentation of skinned bodies and quantifiable physiological responses is presented in Table 5 to examine the level of connection between these two independent variables. On the BDC Holstein group, a significant link was observed in several viewpoints between B/W skinned pigmentation, such as WRLV and BRLV, and WSST, ATRS, ST, BT, and HTC variables. The rounded B/W-skinned (WBRV and BBRV) also had a significant relationship, but merely with the WSST score. Another notable finding in the BDC group was that black and white pigmentation on the right-lateral viewpoint (WRLV and BRLV) had a significant connection with the ATRS, ST, BT, and HTC marks. Nevertheless, the WDC Holstein class did not have a single criterion with a significant correlation, despite minor output scores being considered at a moderate level according to the correlation class standard, such as WUPV and BUPV with BF, WRLV and BRLV with WSST and ATRS, and WHNV and BHNV with BSST. Altogether, both the BDC and WDC Holstein classes sent a signal that pigmentation in B/W-skinned cattle was irrelevant to the BF, PL, and RW scores. Thus, underlying all that data exposure, the BDC Holstein group possessed greater potential to develop thermotolerant dexterity. Known that cattle adjust their coats and acclimate to their metabolic rates when exposed to persistent heat stress, which increases the evaporative heat loss (EVHL) mechanism when cowshed ambient temperatures hit 36 ºC (Shephard and Maloney, 2023).

Heat stress in warm-blooded creatures is modulated in the brain, specifically in the preoptic region of the pituitary gland (Nagashima, 2006). Several hormones are involved in this situation, including thyroid, estrogen, prolactin, GH, ACTH, progesterone, glucocorticoids, and oxytocin (Beede and Collier, 1986). Still, the cutaneous region influenced the cows' evaporative heat loss (EVHL) regulation dexterity, followed by the heat shock transcription factors (HSF) and heat shock proteins (HSP), as both were identified as genes related to thermotolerant aptitudes (Collier *et al*., 2008). The skin EVHL role depends on the mass of sweat glands, bristles, pigmentation, and the epidermis' vascular system (Collier *et al*., 2008). Not only pigmentation, but the hair extent level also serves as proof of contributing to the thermotolerant aptitude (Davis *et al*., 2016). This topic suggests a connection with disease resistance and immune responses (Tabor *et al*., 2017). The colour characteristics of pigments are incongruous alongside albinism, which is brought on by a frameshift mutation in an enzyme called tyrosin (Schmutz *et al*., 2004). The white pigmentation of the skin coat exhibits greater dexterity in reflecting and transmitting light radiation than other pigments (Da Silva *et al*., 2003). Further, the ambient factors such as wind velocity, Ta, and rH also play a role in the EVHL level (Collier *et al*., 2008).

Comparative test of physiological responses between the B/W classes

The data distribution test for physiological symptoms, categorized by BDC and WDC pigmentation, yielded normally distributed results with a p-value greater than 0.05 in all cases. Following this, the mean-difference comparison analysis generated a surprising result; when the correlation test was executed, several scores were labelled statistically as significant. However, this was not consistently expressed when the t-test was performed between the two classes on the entire gaugeable physiological response or vital symptoms, as shown in Table 6. All variables were perfectly insignificant. Still, if other perspectives were settled, such as the data trends elaborated upon extensively, it would emerge as a splendid and attractive pattern. For example, the BDC Holstein class performed better than the WDC, particularly in terms of the determinable vital symptoms, except for the pulse rate (PL). Pointedly, the BDC Holstein class performs better than WDC on BF (2.23 points) and RW (0.20 degrees) parameters, but the PL sent an odd signal, with WDC being greater than BDC (3.96 points). The vital symptoms of the BDC class score lower than those of the WDC, indicating a lesser risk of experiencing heatstroke or a better thermotolerant aptitude. Individually, extra points above 100 of the HTC score were a valuable signal, as with the uncovered HTC score of the BDC group in Table 6. It enabled the composition of a rational argument from the underlying evidence that the BDC Holstein group had better dexterity in dealing with ambient heat stress.

Table 6. Mean-differences test vital symptoms output of lowland Indonesian cows based on B/W-skinned-dominated categories

Vital symptoms	BDC (Mean ± SE)	WDC (Mean ± SE)
Breathing frequency (BF) (times minute^{-1})	65.14 ± 1.75 ^{ns}	67.37 ± 4.03 ^{ns}
Pulse rate (PL) (times minute^{-1})	87.77 ± 1.82 ^{ns}	83.81 ± 1.98 ^{ns}
Rectum's warmth (RW) (ºC)	38.35 ± 0.06 ^{ns}	38.55 ± 0.22 ^{ns}
White-skinned spot temperature (WSST) (ºC)	33.64 ± 0.09 ^{ns}	33.94 ± 0.19 ^{ns}
Black-skinned spot temperatures (BSST) (ºC)	34.01 ± 0.08 ^{ns}	34.18 ± 0.17 ^{ns}
Average-skinned temperatures (AST) (ºC)	33.82 ± 0.09 ^{ns}	34.06 ± 0.18 ^{ns}
Skin's temperatures (ST) (ºC)	33.35 ± 0.06 ^{ns}	33.40 ± 0.19 ^{ns}
Body's temperatures (BT) (ºC)	37.65 ± 0.05 ^{ns}	37.82 ± 0.20 ^{ns}
Heat tolerance coefficient (HTC)	117.20 ± 1.34 ^{ns}	112.79 ± 5.17 ^{ns}

BDC: black-skinned dominated cows; WDC: white-skinned dominated cows; SE: standard error; ns: insignificant; and identical superscript letters in levelled lines designated insignificant mean differences p>0.05.

A published paper reported that Holstein's dominant white skin coat colouring has a positive effect on productive and reproductive traits (Becerril *et al*., 1993). Linear findings also stated that dozens of dairy cows' distinctive physical characteristics include white skin coat markings (Liu *et al*., 2009). White-skinned cows, which are predominantly white, nursed in a tropical region with controlled ambient conditions and shaded from daily sunlight exposure, presented better performance data on milk yield than black-skinned cows (Maia, da Silva, Bertipaglia, *et al*., 2005). Nevertheless, a handful of scientific counterarguments were also provided by other investigators, such as the fact that significantly yielding Holsteins frequently exhibit significant levels of black skin-coat colouring (Laible *et al*., 2021). Regarding lifespan, Holstein cows wearing 90% black skin-coats outlived those with 90% white skin-coats (Lee *et al*., 2016). An experiment also found that darker-pigmented cattle have a lower epidermal temperature by 1ºC than brighter ones in hot ambient conditions; conversely, the brighter-skinned cattle exhibit greater responses when exposed to cold weather (Lysyk, 2008). Those documentations were consistent with the present exploration findings, even though the spans were not as wide as the score. Splitting data based on divergent classes (BDC and WDC) highlights the fact that BDC Holstein performed well in terms of the expressed vital symptoms.

Conclusion

Classifying lowland Indonesian Holstein cows based on the B/W-pigmentation-dominated skin was a promising approach to increase the heat tolerance coefficient (HTC) score and maintain steady physiological thermoregulation, thereby minimizing the calamitous effects of heatstroke. Deductible, the type of Indonesian Holstein cows that should be extended by smallholder dairy farmers (SDF) in Indonesia's lowland region, was the BDC Holstein type, specifically both lateral sides and the upper area of the body, due to preliminary results manifesting a prominent vital signal than the WDC group. Following up on these topics related to milk yield and persistency traits is warranted.

Conflict of interest

Regarding the current paper, nobody's possible conflicts of interest are disclosed. Each writer had already approved the contents of the text.

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Author's contribution

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