

**Title**

The effect of using different regions of interest on local and mean skin temperature

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## **Abstract**

The dynamic nature of tissue temperature and the subcutaneous properties, such as blood flow, fatness, and metabolic rate, leads to variation in local skin temperature. Therefore, we investigated the effects of using multiple regions of interest when calculating weighted mean skin temperature from four local sites. Twenty-six healthy males completed a single trial in a thermonetural laboratory (mean  $\pm$  SD): 24.0 (1.2) °C; 56 (8%) relative humidity; < 0.1 m/s air speed). Mean skin temperature was calculated from four local sites (neck, scapula, hand and shin) in accordance with International Standards using digital infrared thermography. A 50 x 50 mm square, defined by strips of aluminium tape, created six unique regions of interest, top left quadrant, top right quadrant, bottom left quadrant, bottom right quadrant, centre quadrant and the entire region of interest, at each of the local sites. The largest potential error in weighted mean skin temperature was calculated using a combination of a) the coolest and b) the warmest regions of interest at each of the local sites. Significant differences between the six regions interest were observed at the neck ( $P < 0.01$ ), scapula ( $P < 0.001$ ) and shin ( $P < 0.05$ ); but not at the hand ( $P = 0.482$ ). The largest difference ( $\pm$  SEM) at each site was as follows: neck 0.2 (0.1) °C; scapula 0.2 (0.0) °C; shin 0.1 (0.0) °C and hand 0.1 (0.1) °C. The largest potential error (mean  $\pm$  SD) in weighted mean skin temperature was 0.4 (0.1) °C ( $P < 0.001$ ) and the associated 95% limits of agreement for these differences was 0.2 to 0.5 °C. Although we observed differences in local and mean skin temperature based on the region of interest employed, these differences were minimal and are not considered physiologically meaningful.

## *Keywords*

Tissue temperature; infrared thermography; variability; region of interest; thermoregulation

## 1. Introduction

Skin temperature measurement is commonly used to explore the interaction between human thermophysiology and the external environment (Livingstone et al., 1987). Local skin temperature can be used to monitor and detect inflammation associated with knee replacement (Haidar et al., 2006; Mehra et al., 2005), rheumatoid arthritis (Rajapakse et al., 1981), osteoarthritis (Denoble et al., 2010), allergies (Rokita et al., 2011), frozen shoulder (Vecchio et al., 1992) and even tendinitis (Miyakoshi et al., 1998). Mean skin temperature on the other hand, is most commonly used as an important component of overall thermal stress (Frim et al., 1990), thermal comfort (International Organisation for Standardisation, 2004) and in the calculation of mean body temperature (Colin et al., 1971).

In order to truly measure mean skin temperature accurate measurements must be taken from the body's entire surface area. Obviously this is not possible, resulting in the estimation of mean skin temperature. Therefore, the greater the number of sites the more accurate the estimation (Taylor et al., 2014), although these larger site formula are not without limitations. The calculation of mean skin temperature generally requires the use of 3 to 15 local skin temperature sites with appropriate weightings (Choi et al., 1997). In field use, numerous sites can become time-consuming, cumbersome and restrictive to movement. Olesen (1984) proposed that if environmental conditions produced small intra-site skin temperature variability, such as that seen in hot or thermoneutral conditions, relatively less sites were necessary for accurate mean skin temperature compared to formula needed in cold ambient temperatures. Due to the dynamic nature of skin temperature and the subcutaneous properties altering intra-site variability, such as blood flow, fatness, and metabolic rate, subjective placement of measurement devices could result in erroneous calculations of mean skin temperature, particularly during repeated trials. Unfortunately, current formulae (Hardy et al., 1938; Ramanathan, 1964) and standards (International Organisation for Standardisation, 2004) have failed to provide specific anatomical landmarks for the placement of monitoring devices. This has led to the subjective placement of conductive and infrared devices within a given region of interest (Frim et al., 1990; Pascoe et al., 2012).

Cutaneous blood flow and microcirculation are two factors that are known to influence skin temperature (Maley et al., 2014). Whilst alterations in cutaneous flow alter skin temperature at a gross level, local deviations are more difficult to accurately predict due the complexity of the microvasculature distribution patterns (Braverman, 2000). Indeed, with the complexity of the underlying anatomy of various measurement sites, it is plausible that

variations in the underlying tissue composition including muscle, adipose tissue, blood vessels, bone, tendons, and ligaments could alter skin temperature within a region of interest (Ludwig et al., 2014). It is well established that adipose tissue has insulating properties, which affects thermal conductivity and, by altering heat transfer between the core and skin, influences skin temperature measurement (Cooper and Trezek, 1971). Several studies have demonstrated this by comparing the thermal response of obese and lean individuals (LeBlanc, 1954; Livingstone et al., 1987; Savastano et al., 2009). LeBlanc (1954) showed changes in skin temperature were proportional to the degree of adiposity between individuals and Fournet et al. (2013) found a significant relationship between skin fold thickness and skin temperature distribution at the anterior torso, a region of high adiposity.

Several studies have shown significant variations in skin temperature at local sites, which could affect mean skin temperature calculation (Frim et al., 1990; Livingstone et al., 1987; Pascoe et al., 2012). Both Frim et al. (1990) and Livingstone et al. (1987) reported significant variations in local sites, including the torso, of over 7 °C as measured by thermistors and thermography. However, it is unclear if these known temperature variations in local sites have the potential to propagate to erroneous calculations of mean skin temperature. Therefore, the purpose of the current study was to examine the effects of using multiple regions of interest when calculating mean skin temperature from four local sites in healthy males.

## 2. Methods

### *Participants*

After ethical approval from the Human Research Ethics Committee (Queensland University of Technology), 26 healthy, recreationally active males volunteered to participate in this study (Table 1). Prior to commencing the study all participants completed a health screen questionnaire and informed verbal and written consent was acquired. Contraindications for participation included a history, or current existence of any cardiopulmonary disease, acute skin conditions (e.g. adhesive tape allergy), any metabolic, arterial, venous or lymphatic pathology, current history of smoking, or the use of any medication that alters cardiovascular function or thermoregulation.

**INSERT TABLE 1 HERE**

## Procedure

### *Pre-experimental Protocol.*

Participants were instructed to avoid prolonged sun exposure five days prior to the testing day to prevent sunburn. Where appropriate, any measurement site with exposed hair was shaved at least 36 hours before testing (Togawa and Saito, 1994) to prevent inflammation or damage to the skin surface artificially raising skin temperature and influencing infrared measurements (Merla et al., 2010). In preparation for testing, participants did not engage in exercise, ingest caffeine or alcohol 24 hours prior to (Ammer and Ring, 2006), have a hot shower within two hours of arriving to the laboratory and kept the measurement sites clean of ointments and cosmetics (Selfe et al., 2006).

### *Experimental Protocol*

Initially four body locations (Figure 1) in accordance with ISO 9886 (International Organisation for Standardisation, 2004) were cleaned with alcohol swabs to remove any contaminants influencing skin emissivity (Bernard et al., 2013). The four sites were marked with a square bordered by 3 mm x 50 mm strips of aluminium tape on each side (3M, St. Paul, Minnesota, United States), providing a 50 mm x 50 mm measurement area. The tape acted as an inert marker that allowed for the identification of each region of interest during post-processing of the thermal images (Costello et al., 2012a). The readings from these sites were used to calculate mean skin temperature using the following equation (International Organisation for Standardisation, 2004):

$$\bar{T}_{sk} = (T_{neck} \cdot 0.28) + (T_{scapula} \cdot 0.28) + (T_{hand} \cdot 0.16) + (T_{shin} \cdot 0.28)$$

Where  $T_{neck}$  represents the skin temperature ( $^{\circ}\text{C}$ ) of that region and the numerical value (e.g. 0.28) is the weighted value applied to that site. A conventional seated 20 minute acclimation (Marins et al., 2014) took place in a temperature- controlled, fluorescently lit room without the existence of electric heat generators, wind drafts or external radiation. Skin temperature measurements were taken in a controlled thermoneutral environment;  $24.0 \pm 1.2$   $^{\circ}\text{C}$ ,  $56 \pm 8\%$  relative humidity (QuestTEMP 36, 3M, St. Paul, Minnesota, United States),  $< 0.1$  m/s air speed (Kestrel Pocket Weather 4000, Nielsen-Kellerman, Boothwyn, Pennsylvania, USA). Participants clothing ensemble consisted of shoes, socks, underwear and shorts for the duration of experimental testing.

Data collection consisted of repeated skin temperature measurements taken using a calibrated, uncooled digital infrared camera A305sc (FLIR Systems, Wilsonville, Oregon, USA) with an image resolution of 320 x 240 pixels and a minimal detectable temperature difference of  $<0.05$  °C at 30 °C. The infrared camera was positioned on a level tripod perpendicular to the seated, resting participant (90°) at a distance of 0.8-1.1 m depending on the height and size of the participant (Ammer and Ring, 2006). The camera was assembled and allowed to stabilise for at least 60 minutes prior to subject arrival (Grgić and Pušnik, 2011). A constant skin emissivity for the infrared camera was set to  $\epsilon = 0.98$  in accordance with previous research (Steketee, 1973).

**INSERT FIGURE 1 HERE**

#### *Post processing analysis*

Post-processing FLIR R&D software (version 3.4.13191.2001, FLIR Systems, Wilsonville, Oregon, USA) was used to input recorded variables such as ambient temperature, relative humidity, camera distance and skin emissivity. Skin temperature was derived from three thermal images that encompassed the four regions of interest taken within a 15 second period. Each region of interest was divided into four equal quadrants with a mean  $\pm$  SD pixel size of  $15 \pm 2$  by  $15 \pm 2$ , with an additional equal quadrant placed over the centre of the region (Figure 2). Further to this, skin temperature of the whole region of interest was measured (mean  $\pm$  SD pixel size =  $30 \pm 4$  by  $30 \pm 5$ ).

**INSERT FIGURE 2 HERE**

#### *Statistical analysis*

The data are displayed as mean  $\pm$  SD unless otherwise stated. Normality was assessed using descriptive methods (skewness, outliers, and distribution plots) and inferential statistics (Shapiro–Wilk test). When the assumption of sphericity was violated, significance was adjusted using the Greenhouse-Geisser method to adjust the degrees of freedom to increase the critical values of the F-ratio. Four separate repeated measures analysis of variance (ANOVA) were performed to assess the effect of region of interest (total region of interest [Total], top left quadrant [QLTop], top right quadrant [QRTop], bottom left quadrant [QLBottom], bottom right quadrant [QRBottom], centre quadrant [Centre]) on the four local sites. Pair-wise comparisons, using a Bonferroni correction, were performed where significant differences were observed in region of interest. Mean skin temperatures,

calculated using the coolest and the warmest regions of interest at the four local sites, was established for each participant and analyzed using descriptive statistics and a paired sample t-test. All data was analyzed using SPSS (SPSS version 21.0, SPSS Inc., Chicago, USA). Statistical significance for the ANOVA was set to  $P < 0.05$ .

### 3. Results

Highly significant main effects for region interest was observed at the neck ( $P = 0.009$ ), scapula ( $P < 0.001$ ) and shin ( $P = 0.015$ ); but not at the hand ( $P = 0.482$ ; Figure 3; Table 2). Post hoc differences are displayed in Table 2. The largest difference ( $\pm$  SEM) in local skin temperature between these regions were as follows: neck  $0.2 \pm 0.06$  °C, scapula  $0.2 \pm 0.03$  °C; shin  $0.1 \pm 0.05$  °C and hand  $0.1 \pm 0.08$  °C (Figure 3).

**INSERT FIGURE 3 HERE**

**INSERT TABLE 2 HERE**

Mean skin temperature differed by  $0.4 \pm 0.1$  °C ( $P < 0.001$ ) when calculated using a weighted formula incorporating the minimum and maximum temperature regions of interest at the four local sites. The associated 95% limits of agreement for these differences was 0.2 to 0.5 °C (Table 3).

**INSERT TABLE 3 HERE**

### 4. Discussion

To the best of our knowledge, this is the first investigation to examine the effects of using multiple regions of interest when calculating mean skin temperature from four local sites in healthy males. Despite local and mean skin temperature being popular outcome measures in clinical settings and in applied human physiological research, it is currently unknown how using different regions on interest may alter the measurement. Established thresholds for localised skin temperature differences of 2.2 °C, 2.1 °C and 1.4 °C have been shown to identify risk, severity or even diagnose diabetic foot ulceration (Lavery et al., 2007), complex regional pain syndrome (Wasner et al., 2002), and overuse knee injury (Hildebrandt et al., 2010), respectively. In the current investigation, the region of interest had a significant effect on three of the four local skin temperatures sites, in addition to mean skin temperature. The largest mean difference observed in the current study was 0.2 °C and 0.4

°C, for local and mean skin temperature respectively. As such, the present data indicates that region of interest has limited effect in young males at rest; however, its influence on skin temperature during exercise in high ambient environments warrants further research.

Livingstone et al. (1987) investigated the variation in four torso sites, and calculated mean torso temperature to determine the propagation of error using both thermistors and thermography. Livingstone et al. (1987) found skin temperature variability of up to 7.3 °C across the torso, but poorly defined, non-standardised regions of interest meant that reported variations would be of little practical value for the calculation of mean skin temperature. Frim et al. (1990) investigated variation at four torso sites using five thermistors at each site. Each site consisted of a central thermistor, with 4 other thermistors (above, below, left and right) placed 2.5 cm away from the central thermistor. Although mean ranges (difference between warmest and coolest thermistor at each site) were between 1 to 3 °C depending on the ambient temperature, authors also noted that variations could be up to 7 °C between individuals in identical conditions. The magnitude of these variations significantly exceeds variations found in the current study. However, the use of thermistors in both of these studies carries several limitations influencing variability, including the pressure at which the device is applied (Stoll and Hardy, 1950), the microenvironment created by the contact point (Buono and Ulrich, 1998; Tyler, 2011) and the fact that each measurement is merely a spot reading which would be unlikely to reflect the average temperature of the area (Costello et al., 2012b). Furthermore, both Livingstone et al. (1987) and Frim et al. (1990) conducted investigations using only torso sites. Torso sites have a high variability in subcutaneous adipose tissue distribution and given the role of adiposity in skin temperature (LeBlanc, 1954; Livingstone et al., 1987; Savastano et al., 2009), it would be expected that this demonstrated variability might not apply to other sites. Whilst many weighted equations for mean skin temperature calculation do indeed contain at least one of the selected torso sites (Choi et al., 1997) they also contain other more peripheral sites. In the current investigation, the hand was the only site not to reflect significant differences within the site. It is logical to expect a peripheral site with relatively superficial vasculature, bone and muscle, such as the hand, to show large intra-site variability. Recently, Pascoe et al. (2012) demonstrated that the degree of variability ranged dramatically between the peripheral skin temperature sites. However, the lack of adiposity and minimal convexity of the hand relative to the other three sites may have resulted in a more uniform skin temperature measured with infrared thermography. Temperature error associated with viewing angle and convexity of an object

have been defined previously (Cheng et al., 2012). Cheng et al. (2012) reported angular deviations of a camera between 40 and 60° against a flat surface and begin to subtly limit the radiative energy measured by the camera, resulting in errors of up to 0.3 °C. The convex nature of the human form influences the true emissivity of a given measurement site, that can result in errors of  $\pm 2$  °C if the viewing angle exceeds 70 to 80° (Cheng et al., 2012). However, the controlled viewing angle in the present study, in addition to the use of regions of interest on surfaces of relatively little convexity, means that reflected emissivity related error was minimal.

The greatest difference observed in the current study was 0.2 °C for local, and 0.4 °C for mean skin temperature. Despite showing statistical significance (most likely a product of the power of the sample) and some research suggesting these values may be meaningful (Vardasca et al., 2007), each fall within the proposed clinically significant threshold for healthy normalised skin temperature differences of 0.5 °C (Selfe et al., 2008). Interestingly, Pascoe et al. (2012) used infrared thermography to investigate skin temperature variability at 13 sites and attempted to quantify the error in calculating mean skin temperature in environmental conditions of 30 °C, 40% relative humidity. The average range of skin temperature at each site was  $3.19 \pm 0.93$  °C, and for all prediction equations used, mean skin temperature ranged over 3 °C from using the coldest and hottest temperature at each site. However, several methodological problems may be able to explain the contrary results to that reported in the current study. Firstly, the variability in local and mean temperature was conducted using the coldest and hottest temperature at each location. Whilst this was not clearly described, this suggests that researchers used the hottest and coldest single pixel within the area of interest. The use of these values is practically unreasonable as no contact device could measure an area of that small a size. Contrary to this method, the present study used the hottest and coldest average temperature region (25 mm<sup>2</sup>) within an appropriately sized local site (50 mm<sup>2</sup>), meaning that the total area encompassed within each region was similar to the measurement area of a contact device (Harper Smith et al., 2010). This methodology resulted in minimal changes in overall mean skin temperature. In addition to this, the demonstration of the regions of interest by Pascoe et al. (2012) suggests two further major limitations. The first is that the regions of interest are quite large, meaning they encompass areas that are not practical for the placement of contact devices. Secondly, the illustration of subject positioning suggests that some portions of the regions of interest were observed at considerable oblique angles to the camera due to the curvature across large skin

sites. Subsequently, measurement errors may have been exacerbated due to the reflected emissivity at the skin surface (Cheng et al., 2012). This introduction of error greatly increases the apparent variability in skin temperature.

The findings of this study are limited to tightly controlled laboratory conditions for young healthy males. Future studies should therefore investigate skin temperature variability in different populations, including females, elderly and injured groups. We were able to rule out erroneous translation of flexible local sites to mean skin temperature during resting, thermoneutral conditions. However, variability and therefore potential error within localised skin sites as a consequence of metabolic heat production and environmental manipulation requires further research. Moreover, the present study used 4-sites commonly used for the calculation of mean skin temperature. It is currently unknown whether the use of other equations, and other local skin temperature sites, will affect the propagation of error into mean skin temperature. Finally, investigation of variability within local skin temperature sites such as the knee, shoulder, elbows and ankles may be of interest due to clinical implications.

## **5. Conclusion**

Despite finding highly significant differences in neck, scapula and shin, the magnitude of variation at each of these sites was negligible ( $<0.3$  °C) in healthy young males at rest in a thermoneutral environment. Furthermore, these variations did not introduce a clinically meaningful error ( $<0.5$  °C) in mean skin temperature. However, further research examining the influence of using different regions of interest to calculate skin temperature during exercise in high ambient environments is warranted.

## **Vitae**

**Nirav Maniar** has completed a Bachelor of Exercise and Movement Science at Queensland University of Technology. He is currently completing a Masters by Research in the School of Exercise Science at Australian Catholic University in Melbourne. He has completed work in thermal physiology and has current interests in injury prevention and rehabilitation.

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