THERMOGRAPHIC IMAGING OF THE PREGNANT UTERUS: THE EVALUATION OF A NOVEL DEVICE AND ITS USE IN THE ASSESSMENT OF INTRA-UTERINE CHARACTERISTICS DURING PREGNANCY

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A Dissertation presented to the Faculty of Medicine and Surgery at the University of Malta, as part fulfilment of the requirements for the Doctor of Philosophy Degree

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Dedication

To my husband who continues to support me through it all.

To my parents who amongst other things, taught me the value of an education.

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I would like to thank the 'three pillars' who selflessly provided support and advice throughout this long journey, and dedicated much time of their own towards a project which at times looked too big to handle – Mark, Lisa, Dad, I will forever be grateful!

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List of Abbreviations

AAT = American Association of Thermography

AC = Abdominal circumference

AFI = Amniotic Fluid Index

ALARA = As Low as Reasonably Achievable

BMI = Body Mass Index

CT = Computed Tomography

CTPA = Computed Tomography Pulmonary Angiogram

DSP = Digital Signal Processing

DVP =Deepest Vertical Pool

EM = Electromagnetic Radiation

FDA = Food and Drug Administration

FPA = Focal Plane Array

Gy = Greys

ICC = Intra-class Correlation Coefficient

IR = Infrared

IRT = Infrared Thermography

IUGR = Intrauterine Growth Retardation

IVF = In-vitro Fertilisation

LLQ =Left Lower Quadrant

LUQ = Left Upper Quadrant

LW = Longwave

MHRA = Medicines and Healthcare Products Authority

MRI = Magnetic Resonance Imaging

MW = Mediumwave

NOIS = National Obstetric Information System

NSAID's = Non-Steroidal Anti-Inflammatory Drugs

O&G = Obstetrics and Gynaecology

OHA = Oral Hypoglycaemic Agent

PlGF = Placental Growth Factor

RLQ = Right Lower Quadrant

ROI = Region of Interest

RUQ = Right Upper Quadrant

SW = Shortwave

T_{amb} = Ambient Temperature

T_b = Body Temperature

$T_{lowCrit}$ = Lower Critical Zone

T_{upCrit} = Upper Critical Zone

V/Q = Ventilation Perfusion Scan

VEGF = Vascular Endothelial Growth Factor

Abstract

In humans, heat production and heat dissipation is regulated by metabolic and vascular mechanisms which maintain a delicate equilibrium between the body temperature and the surrounding environmental temperature. In pregnancy, an increase in maternal metabolic rate and heat dissipation from the foeto-placental unit are added factors resulting in heat production. Infrared thermography is a technique to record the naturally emitted infrared radiation from the area studied, making it a safe, non-invasive, non-contact device which can be used to evaluate the temperature patterns over the pregnant abdomen.

The principle goal of this study was to assess the use of thermographic technology to acquire abdominal skin temperature data and explore if this can be related to ultrasound imaging measurements in pregnant women. The research was divided into four stages: (i) a preliminary pilot study to empirically assess the feasibility of taking thermographic measurements and correlating them with the deepest liquor pool measured on ultrasound. This was conducted on 46 patients and provided a basis for adjustments and refining of the methodology used in the second phase of the study; (ii) a main study which investigated a cohort of 42 healthy pregnant women of gestational age between 28 and 31 weeks. The objective was to investigate the presence and quantify any relationship between the temperature readings on thermographic imaging and the liquor pool measurements on ultrasound. The effect of maternal and foetal factors on this relationship was also analysed. Furthermore the time profile for acclimatisation of these patients to ambient temperature was explored; (iii) a second study investigated a group of 10 patients suffering from diabetes during pregnancy. The same relationships between thermography data and ultrasound measurements as in the main study were investigated together with acclimatisation profiles of these patients. Results from this cohort and the non-diabetic cohort were compared; (iv) an intra-rater and test-retest reliability study was conducted on the methodology of using the thermographic software to process image data in this obstetrics application.

The results showed positive correlations between the temperature measurements carried out by thermography and the liquor pool depths on ultrasound in both healthy and diabetic patients. The application of cream on the abdominal surface was found to be a factor which affects this relationship in both groups, whilst the foetal abdominal circumference was found to have an effect in the diabetic group. The acclimatisation period required for the two groups was determined by analysing the statistical significance of the relationship over time points. A predicative model based on this relationship was generated for both groups. Both reliability studies showed excellent levels of agreement in the method of processing thermographic data by different users and by the same operator during distinct study days.

The motivation for this research undertaken was to assess if thermography can be considered as a useful modality in obstetrics in humans. The study provided new insights into the way this device can potentially be used in this medical field, and makes an original contribution to the research on medical thermography by providing an added understanding to the way thermographic data can be interpreted when paralleled with conventional methods of analysis such as ultrasound.

1 Introduction

1.1 Literature Review

1.1.1 Physiology of Thermoregulation

Humans are a homoeothermic species, meaning that the body core temperature is maintained throughout a variety of ambient temperatures, allowing them to be active in different environments. Thus, this gives humans an advantage over other non-homoeothermic species (Kanosue, Crawshaw, Nagashima, & Yoda, 2010).

Heat is the main by-product of all metabolic processes that constantly occur within the human body. It is therefore vital that there be a control system which can maintain a near constant temperature within the body allowing vital organs, in particular metabolic and enzymatic reactions, to continue to function despite fluctuations of ambient temperature. In order to function the control system, known as thermoregulation, utilises a complex system of neural, vascular and chemotactic mechanisms amongst others. The temperature required for the body to function at a cellular level is called the core temperature and averages between 36.6˚C and 37˚C (Hall, 2006). Various enzyme systems have a narrow temperature range within which their function is optimal. Also, the speed of the chemical reactions within the body can change secondary to body temperature. Normal stable bodily function is therefore highly dependent on the relative consistency of body temperature (Barrett, K. E., & Ganong, 2012).

Thermoregulation is the balance of heat production and heat dissipation – the delicate equilibrium between the body temperature and the surrounding environmental temperature. When heat produced within the body is greater than the heat lost, it builds up and the body temperature rises as a result. This triggers mechanisms of heat loss through the peripheral body regions, mainly the skin. Heat is also lost during exhalation of pulmonary gases. Conversely, when the heat loss surpasses that being produced, the body temperature declines and the reaction of the body is to maintain and preserve as much heat as possible (Hall,

2006). When the body temperature is elevated, the posterior hypothalamus inhibits the sympathetic vasoconstriction of blood vessels, resulting in vasodilatation, which enables more efficient heat transfer from the core to the peripheral areas of the body as shown in Figure 1. There is also a decrease in heat production by inhibiting processes such as chemical thermogenesis. In addition to this, sweating may be triggered in order to increase the rate of heat loss from the body surface by evaporation. On the other hand, when the body is cold, the sympathetic centres in the posterior hypothalamus are stimulated and vasoconstriction ensues. This decreases the blood flow to the peripheries and conserves core heat. Other methods of heat preservation include increasing heat production, by for example shivering and non-shivering thermogenesis (Hall, 2006).

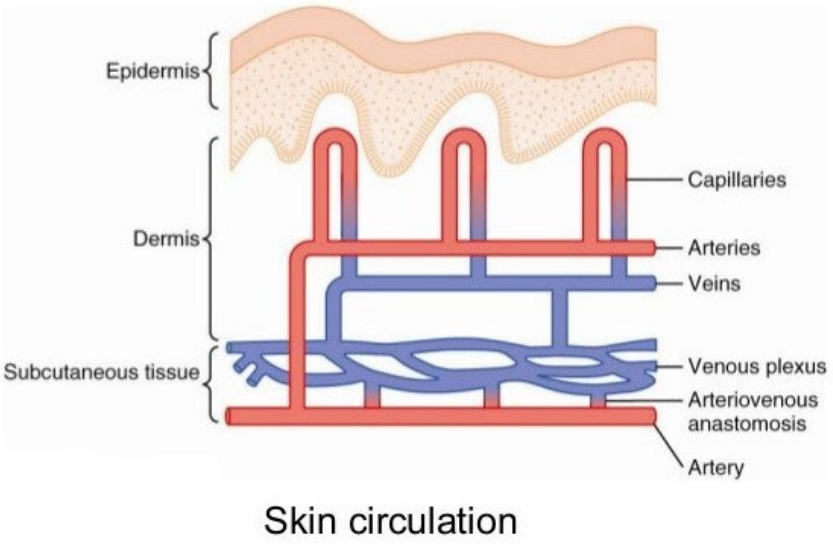


Figure 1. Skin circulation at the skin surface

(Hall, 2006)

The main methods of heat dissipation through the body surface is radiation and conduction, followed by evaporation at normal room temperature (Hall, 2006) (see Figure 2). This is known as sensible heat loss. Radiation is the main way heat leaves the surface of the human body, and is on the infra-red spectrum of radiation at a wavelength between 5 and

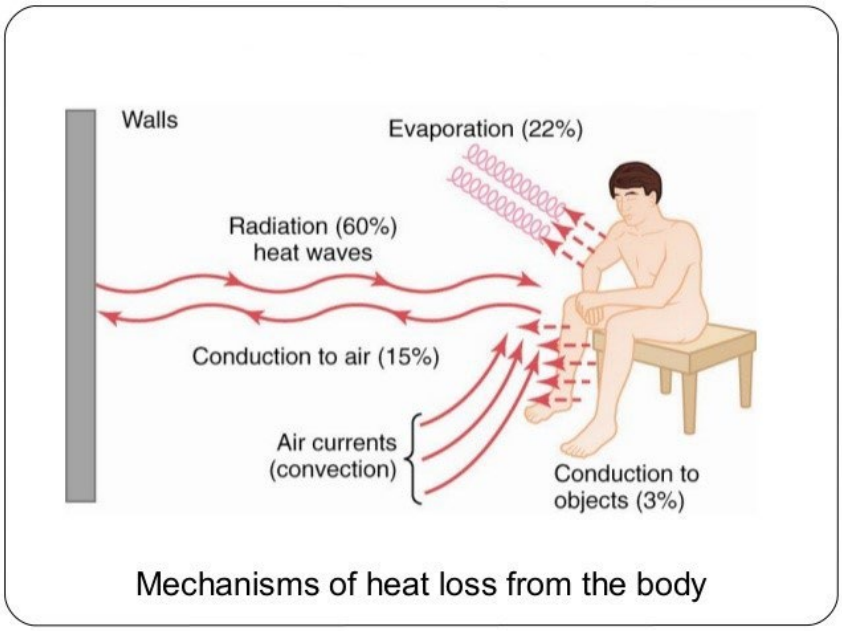


Figure 2. Image showing heat dissipation from the body

(Hall, 2006)

20µm. Heat is also lost from the skin via conduction, by transferring of heat by kinetic energy of the surrounding air particles. It is important to note that once the surrounding air reaches the same temperature as that on the body surface, there will be no further loss of heat via conduction. The third method of heat dissipation is by evaporation from the skin surface. This is an important method at extreme temperatures when the action of the production of sweat causes the transfer of heat from the body surface to the surrounding environment (Hall, 2006).

An important concept in thermoregulation in humans is the Thermal Neutral Zone. This is defined as 'the range of ambient temperatures without regulatory changes in metabolic heat production or evaporative heat loss' (Boris Kingma, Arjan Frijns, 2012). In other words, it is the range of thermal environment where body temperature is maintained only by dry heat losses. As the ambient temperature (T_{amb}) decreases, the temperature reaches the so called Lower Critical Zone ($T_{lowCrit}$). Here, the basal metabolic rate and body temperature (T_b) are equally matched. Below the level of $T_{lowCrit}$, there is an increase in thermogenesis and a redirection of blood flow from the peripheries to the central body area.

By shivering, the metabolic heat generation increases linearly with decreasing T_{amb} thus retaining thermal balance. As the T_{amb} increases however, and once the temperature goes beyond the Upper Critical Zone (T_{upCrit}), a decrease in thermogenic processes is triggered and may also cause surface evaporation and other processes which necessitate an increase in metabolism. This relationship between metabolism and ambient temperature is demonstrated in Figure 3. The linear slope of this correlation is influenced by the degree of body insulation where more insulation leads to a decline in gradient and a consequent negative shift in $T_{lowCrit}$ (Boris Kingma, Arjan Frijns, 2012).

One aspect of thermoregulation physiology of interest that has been studied extensively is its function in pregnancy, both in humans and animal studies.

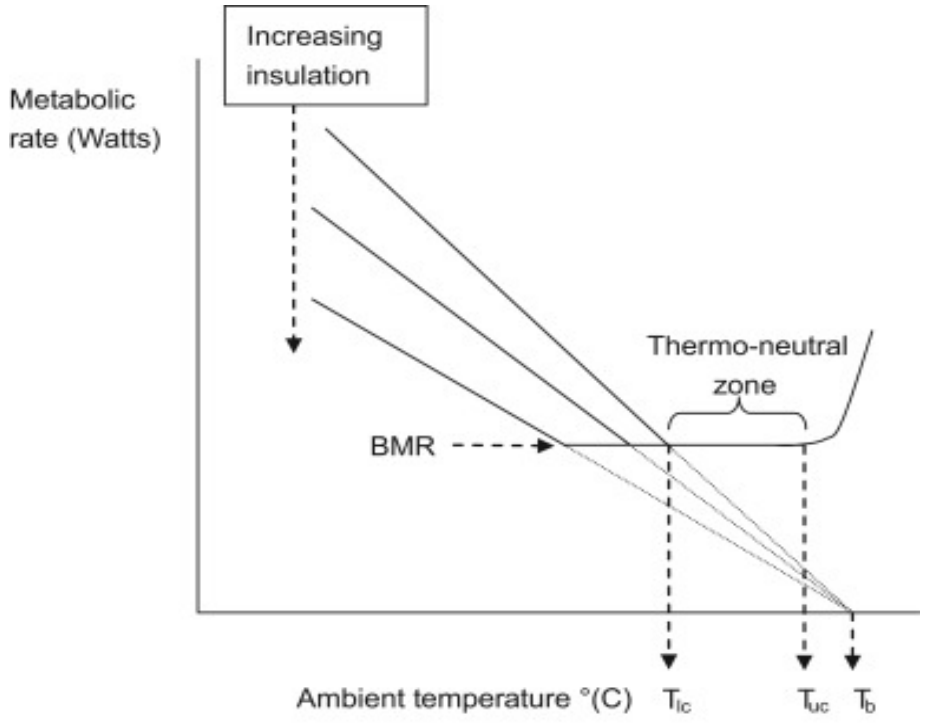


Figure 3. Metabolic rate and the ambient temperature

(Boris Kingma, Arjan Frijns, 2012)

1.1.2 Thermoregulation during pregnancy

It is well known that basal body temperature fluctuations are present during the menstrual cycle, and is in fact a means of natural family planning (Figure 4) (Barrett, K. E., & Ganong, 2012).

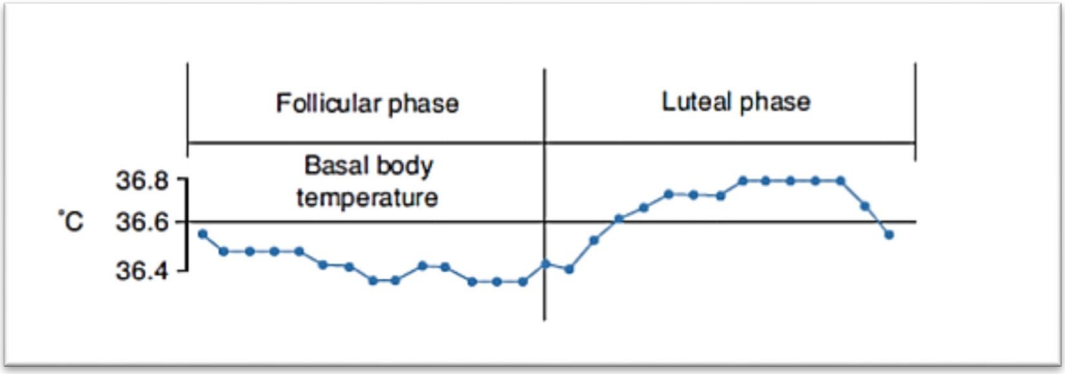


Figure 4. Basal body temperature changes during the menstrual cycle (Barrett, K. E., & Ganong, 2012)

The physiology of the body is significantly altered during pregnancy due to hormonal and biological modifications (Soma-Pillay, Nelson-Piercy, Tolppanen, & Mebazaa, 2016). Physiological changes in the vascular and endocrine system have an effect on the thermoregulatory function during pregnancy. There are mainly two sources of increased heat production that are happening during pregnancy: an increase in maternal metabolic rate as well as the added heat dissipation from the foetal-placental unit.

Most of the animal studies carried out to establish the dynamics of heat transfer within the foetal-placental unit, have been performed on sheep (D. Gilbert, Power, & Fetaland, 1986; Gordon & Gunn, 1987; Schröder & Power, 1997; Yoneyama, & Sawa, 1998; Helen P. Laburn et al., 2002; H. P. Laburn et al., 1992; Kubonoya et al., 1998). The temperature difference between the foetus and the maternal sheep was shown to be 0.6 ˚C in neutral ambient temperatures. This temperature gradient seems to reach stability in the region of ≥125 days of gestation, and despite the normal maternal temperature fluctuations that occur in the last few weeks of gestation, the gradient seems to maintain stability (H. P. Laburn et

al., 1992). Power et al., (1987) looked at the effect of cooling on the thermoregulatory mechanisms in the foetus. By inserting coil tubing around the trunk of the foetus and running tap water through it, the temperature was dropped to 2 degrees for up to 120 minutes. The foetal temperature was seen to fall rapidly when compared to the maternal temperature. This reduction in temperature resulted in a drop of oxygenation and increased acidosis. There was also a failure of triggering shivering and non-shivering thermogenesis. Interestingly, as the foetus was perfused and oxygen levels increased, a modest effect of non-shivering thermogenesis was seen, leading to an increase in free fatty acid and glycerol plasma concentration. The foetal temperature increased even further when the cord was occluded. This demonstrated a clear relationship between the oxygenation levels and the thermoregulatory mechanisms in the foetus, and how these mechanisms are limited by oxygenation of brown fat and augmented by cord occlusion (Power et al., 1987). In another study, an increase in foetal temperature of 0.12 ˚C above baseline, was demonstrated during serial cord occlusion (Kubonoya et al., 1998) The route of heat loss in foetal lambs was shown to be mainly via the placenta (84.5%) but also, through the foetal skin, the surrounding amniotic fluid and the uterine wall (15%) (R. D. Gilbert, Schroder, Kawamura, Dale, & Power, 1985). These studies demonstrated that not only is the foetal thermoregulation integrally linked to the oxygenation of the foetus, but also that the route of heat loss in not exclusively via the placental unit.

These studies were conducted in animals, however, our present research questions thermoregulatory mechanisms in humans. The invasive methods used to carry out these studies cannot be replicated in humans. However, the way thermoregulatory changes occur in pregnancy has been studied extensively using non-invasive methods.

In their study, Hartgill, Bergersen, & Pirhonen (2011), described their work in exploring the pattern of temperature changes during normal pregnancy in humans, where they

explored the relationship between the maternal core temperature and the thermo-neutral zone by using an electronic tympanic probe for maternal temperature and an electronic thermistor probe for the measurement of the room temperature . They carried out a longitudinal study where they explored temperature patterns through 15 normal pregnancies, from week 8 of gestation up to week 52 post-partum. They found that the maternal T_b gradually fell during the pregnancy and remained below the non-pregnant values for up to 3 months' post-partum. The T_{amb} required to reach the thermo-neutral zone was found to decrease from week 8 down to its lowest level at week 36, being 4˚C lower when compared to early pregnancy levels. Their method of measuring tympanic temperature made their application less invasive than previous studies were oesophageal and rectal probes were used (Hartgill et al., 2011).

The way the foeto-maternal unit is affected by temperature during pregnancy has been shown in these studies (Hartgill et al., 2011), however by understanding the development of the placenta and the various pathologies pertaining to the foeto-maternal unit, the use of temperature as a parameter for assessment in pregnancy needs to be understood.

1.1.3 The Anatomy of the Foetal-Maternal unit

In the following section, the placental development and the way it functions within the foeto-maternal unit will be described. Pathologies pertaining to issues with this interface will also be highlighted. This background knowledge must be discussed in order to appreciate the importance of such a structure and how any developmental anomalies may result in errors of function which can be detected on ultrasound.

1.1.3.1 Placental Development

The placenta is an essential interface between maternal and foetal circulations. Its function has an integral role in preventing rejection of the foetus, ensuring adequate respiratory gas exchange and enabling the transfer of foetal toxic metabolic waste into the maternal blood circulation for elimination. The placenta is also important in producing

steroid and protein hormones for the protection of the pregnancy. Once the cellular mass (morula) is formed on day 4 after conception, the cells continue to replicate to form the blastocyst. This then divides into the inner cell mass, the trophoblast and the blastocoele. The amnion develops as a layer from the inner cell mass separating the latter from the blastocoele. Once implantation occurs between day 5 to day 8, the circulation from the endometrium supplies oxygen and metabolic substances to allow the development of the embryo. The trophoblastic layer develops into the cytotrophoblast which migrates deep in the decidual layer. This then proliferates to then differentiate into the syncytiotrophoblast on the maternal side of the placenta. Between these two layers of cells, vascular lacunae form (Edmonds, 2008).

In week 5 of development, the villi continue to form and embed further within the maternal endometrium. This happens whilst the spiral arterioles continue to develop by replacing the vascular muscle and thus changing from narrow arteries to dilated ones. This allows lower resistance within the vessels in order to permit better gas transfer, exchange of nutrients and removal of waste products (Edmonds, 2008) (Figure 5).

1.1.3.2 Pathologies of Placental Function

The physiological mechanism for the developmental abnormalities in the placental vasculature which lead to obstetric conditions such as pre-eclampsia and intrauterine growth retardation (IUGR) have been the source of many studies. The proposed mechanism is that the spiral arterioles persist with a narrow lumen as a result of defective syncytiotrophoblastic invasion. The narrow lumen then results in an abnormal blood supply to the placenta. Also the vessels may be infiltrated with lipophages and necrosis and thus furthermore reducing the function of the placenta (Pijnenborg, Vercruysse, & Hanssens, 2006).

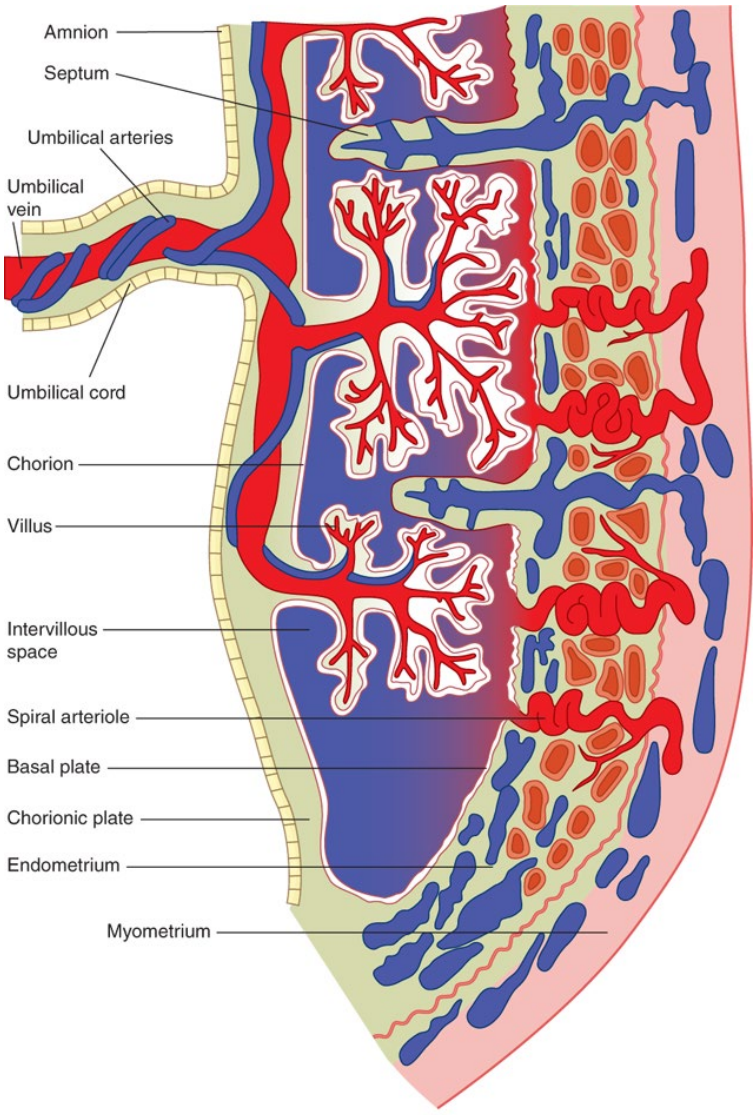


Figure 5. Foetal-placental circulation interface

(Barrett, K. E., & Ganong, 2012)

Poor remodelling and development happens between week 8 and 18 of gestation and leads to what is known as oxidative stress of the villi. This is a state in which there is an excessive accumulation of reactive oxygen which then in turn has a negative effect on the tissues, specifically the villi (Mistry, Kurlak, & Broughton Pipkin, 2013). This syncytiotrophoblast stress can be demonstrated by angiogenic factors such as VEGF (Vascular endothelial growth factor) and PlGF (Placental Growth Factor). In normal pregnancies, the VEGF levels remain constant and the PlGF increases up to approximately 30 weeks. After this, the PlGF starts to fall whilst the VEGF levels start to increase. This is a

similar pattern in cases of late onset pre-eclampsia, meaning that it is difficult to distinguish patients with late onset pre-eclampsia from normal pregnancies solely on the basis on angiogenic factors. In normal pregnancy the terminal villi expand at term in order to continue placental growth. It is believed however, that this growth results in reduced placental perfusion, which may cause the syncytiotrophoblast stress (Staff, Dechend, & Redman, 2013). Ultrasound is another method which can be used to monitor and detect placental changes in pathologies such as pre-eclampsia. This will be discussed further in section 1.1.4.1.

Another condition in pregnancy which has been extensively studied in relation to placental changes is foetal growth. Growth restriction implies that there is a pathological cause for the failure to reach the growth potential of the foetus (Royal College of Obstetricians and Gynaecologists, 2013). There is a distinction between a small for gestational age foetus and a growth restricted foetus. The former is one where the growth is below the 10^{th} centile of a customised growth chart. In the case of small for gestational age foetus, the usual cause is constitutional and is associated with normal umbilical Doppler studies. A growth restricted foetus is one where the reduction in growth rate is due to placental dysfunction (Royal College of Obstetricians and Gynaecologists, 2013). This is subdivided into early and late intra-uterine growth restriction. Early growth restriction is defined as occurring before 34weeks gestation and classically demonstrates abnormal Doppler flow in the umbilical artery as well as foetal compensatory mechanisms.

Baschat (2011) described three phases in the deterioration of the foetal condition in the case of IUGR – the pre-clinical phase, the clinical phase and the deterioration phase. The pre-clinical phase is one where the umbilical flow is reduced due to pathology, resulting in the foetus compensating at best by redistributing blood flow to the heart and other essential organs. The result is a reduced blood flow to the liver which means that there is less glycogen storage and nutrient break down to sustain growth. The redistribution of circulation and

altered nutrient metabolism results in a reduction of abdominal circumference on ultrasound and is seen on the growth chart as declining to below the 10th centile. The clinical stage is when the placental dysfunction is then detected on ultrasound. At this stage there is a degree of arterial redistribution secondary to the increased ventricular afterload. Some blood from the descending aorta will reverse back to the cerebral circulation through the aortic isthmus, thus again redistributing the circulation to more 'essential' organs. The umbilical artery Doppler indices rise at this stage whilst the middle cerebral artery Doppler indices may decrease. The foetus may start to develop a degree of hypoxemia at this stage (Baschat, 2011).

Foetal acidaemia rather than hypoxaemia has a greater risk for developmental delay (Soothill et al., 1992) and this occurs in the deterioration stage. It is here where the foetus finds it difficult to compensate with the reduction in metabolic by-products and the acidaemia.

Baschat (2011) has described the Doppler finding demonstrating differences between early and late onset IUGR. In early onset IUGR, the deterioration occurs when the umbilical and the ductus venosus Doppler progresses from an elevated index to an absent or reversed end diastolic velocity. This stage correlates with deterioration in biophysical profile as well as liquor volume. In late onset IUGR, the umbilical Doppler may be mildly or not at all elevated with no change in the biophysical profile or the liquor volume.

Conditions such as pre-eclampsia and IUGR are routinely investigated by ultrasound in the field of obstetrics. In the previous section, temperature was shown to be an important biophysical factor in the foeto-maternal unit. Temperature acts as a good marker of the metabolic activity occurring within the body, and although we are aware of the presence of the thermo-neutral zone, any significant pathology will push temperatures beyond the T_{upCrit} and the $T_{lowCrit.}$ This makes temperature a feature worth exploring when it comes to a novel method of detection of pathology in obstetrics. Although temperature has been investigated,

this has been done mainly in the setting of maternal exercise (The American College of Obstetricians, 1994; Yeo, 1994) and foetal defects (Edwards, 1986; Milunsky et al., 1992). These findings however, have rarely been translated by investigating a new technique of measuring the temperatures in pregnancy (T. Pereira, Nogueira-Silva, & Simoes, 2016; Simoes, Vardasca, & Nogueira-Silva, 2012), and how this parameter can be used in addition to conventional methods to help in the detection of pathologies such as pre-eclampsia or IUGR.

The present research explores the use of a non-invasive methodology called thermography as a new technique in measuring temperatures in pregnancy, and how this is related to parameters measured by ultrasound.

1.1.4 Non Invasive Diagnostic Imaging in Pregnancy

The options of diagnostic imaging in pregnancy are limited by the safety profile to the foetus. For this reason, the various modalities for imaging are used for different purposes and to varying degrees. The use of ionizing radiation (X-rays and CT scans), Gamma Radiation, MRI and Ultrasound in pregnancy, as well as their safety profiles, will be discussed later.

Ionizing radiation may have use in pregnancy in determining maternal pathology. The strength of this radiation is measured in Greys (Gy). The effect on the foetus includes miscarriage/stillbirth, growth retardation, congenital malformations, neural tube defects including microcephaly and mental retardation. The level of ionizing radiation beyond which major congenital malformations are more likely, was found to be 0.2Gy (Brent, 2006). The gestational age would also make the foetus more susceptible to various effects, with the pre-implantation period (day 0-9) being the highest risk for foetal loss, the organogenesis phase (week 2-8 and 8-15) being of an increased risk for congenital malformations and neural tube defects respectively, and the foetal phase (week 15 onwards) to have the least sensitivity to radiation mainly displaying as mental retardation (Kusama & Ota, 2002).

X-rays are mainly used to investigate maternal pathology for example as chest X-rays or skull X-rays. When administered, they are not directed at the foetus, reducing exposure. Also, the radiation dose used is that of 0.01mGy, which is markedly below the threshold of 0.2Gy. Computed Tomography (CT) is another form of ionizing radiation and may be used in cases of abdominal pain in pregnancy, were a pelvic CT is carried out. This is targeted at the foetus and would have a radiation dose of 20-80mGy (Kusama & Ota, 2002).

Maternal pulmonary embolus investigation is a widely carried out procedure in pregnancy if suspicion arises. The two options for imaging would be a CTPA (Computer Tomography Pulmonary Angiogram) or a V/Q scan (Ventilation/Perfusion Scan). The former uses ionizing radiation and the latter utilises gamma radiation. It was found that although the radiation exposure varied during the three trimesters, the ionizing exposure was overall less to the foetus than the gamma radiation (Winer-Muram et al., 2002). The overall exposure to the foetus is still within minimal amounts for both the modalities and so pose little risk to the foetus (Eskandar, Eckford, & Watkinson, 2010). The main issue with these forms of radiation is the use of radioactive tracers such as iodine or technetium, which could cross the placenta and have potential to affect the foetus. Specifically in the use in maternal thyroid investigation, the radioactive iodine may affect the foetal thyroid if given at 10-12 weeks' gestation (American College of Obstetricians and Gynecologists, 2020).

MRI is another modality that is used for maternal investigation and is gaining popularity for the investigation of foetal conditions such as neural tube defects, thoracic defects, genitourinary defects, and musculoskeletal defects. Magnetic resonance imaging (MRI) aligns the hydrogen atoms and protons in the tissue using magnetic potential, to then apply radiofrequency pulses which will alter this alignment and produce an image. Since there is no ionizing radiation being used here, the posed risk to the foetus is negligible. Some animal models showed a reduced survival rate in mice once exposed at the pre-implanation stage and the recommendation of the MHRA (Medicines and Healthcare Products

Regulatory) is for the avoidance of the use of MRI in the first trimester unless the benefit of such an investigation outweighs the risk (Story, 2015; Narra, Howell, Goddu, & Rao, 1996). The use of MRI in the 2nd and 3rd trimesters is however considered to be safe.

Ultrasound is the main modality used in pregnancy. It is used extensively over all three trimesters as 2D as well as 3D modalities. It has become an integral part of antenatal care and it is an essential investigation tool in congenital anomalies, foetal growth restriction, hypertensive disorders in pregnancy to name a few. Some potential foetal effects have been highlighted, namely thermal injury and mechanical injury. Thermal injury is of more concern especially at early gestations – less than 10weeks (Royal College of Obstetricians and Gynaecologists, 2015). The thermal index is a measure of the ultrasound beam thermal effect which is displayed by the ultrasound machine. The thermal index can potentially increase the temperature of soft tissue and bone by up to one degree. This may cause heat damage to the embryo. It is therefore very important to follow the ALARA principle which stands for 'as low as reasonably achievable' (Royal College of Obstetricians and Gynaecologists, 2015). It has been defined as a recommended protective measure for the subject that is being assessed.

1.1.4.1 The role of ultrasound Doppler in Pre-eclampsia and IUGR

There have been various studies carried out to assess the use of Doppler in combination with biochemical markers in order to predict the risk of pre-eclampsia and triaging the patients into high risk and low risk groups. Odibo et al. (2011) included 447 pregnant women and tested PAPP-A levels, PP13 levels and uterine artery Doppler pulsatility index. Pre-eclampsia was diagnosed in 42 patients and the remaining patients were used as controls. The study showed that early PP13 and PAPP-A levels in the first trimester act as predictors of pre-eclampsia development in the second trimester. An elevated uterine Doppler pulsatility index in the first trimester also corresponds to an increased risk of development of the condition in the second trimester (Odibo et al., 2011). Research has shown that by using

the uterine artery Doppler in the first trimester, it is expected that 81% of women are found to be at risk of early onset pre-eclampsia, 45% of women at risk of late onset pre-eclampsia while 50% of women are at risk of gestational hypertension with a type 1 error rate of 10% (Mone, McAuliffe, & Ong, 2015). Schulman & Fleisher (1986) included 12 non pregnant and 79 pregnant women and carried out serial measurements of the uterine artery Doppler from the last menstrual cycle all through 40 weeks' gestation. They described the early diastolic notch in the Doppler waveform which was present in the non-pregnant women, whilst in the pregnant women, the vessel compliance varied from early pregnancy to 26weeks gestation. Fleisher, Schulman, Farmakides, Bracero, & Grunfield (1986) carried out an additional study looking at 71 patients with hypertensive disease and the Doppler velocimetry of the uterine arteries. They found that in those patients with a systolic to diastolic Doppler ratio less than or equal to 2.6, the pregnancy will not have any complications. If, on the other hand, the ratio is more than 2.6, various complications including stillbirth, intrauterine growth retardation or eclampsia, can occur. This is however an old study and might in itself pose a limitation towards the results described. The use of Doppler studies in both first and second trimester as a form of screening was found to be the most accurate when not associated with other markers such as placental biomarkers. This method was found to have a 10% false positive rate, with the ability to forecast early onset pre-eclampsia in 81%, late-onset in 45% and gestational hypertension in 50% of cases respectively (Bruno, 1994; Mone et al., 2015).

Overall, the role of Doppler studies is to stratify the obstetric population into low risk or high risk for the development of the condition. A guideline has been issued by the Royal College of Obstetricians and Gynaecologists regarding the investigation and management of the small-for-gestational-age foetus. They state that all patients should be risk assessed at the start of the pregnancy and those found to have three or more minor risk factors should have a uterine artery Doppler carried out at 20-24weeks gestation. Of these, those who are found to have an abnormal Doppler, meaning a pulsatility index over the 95th centile, with or without

notching, should have serial ultrasounds starting from 26 to 28weeks gestation to look at umbilical artery Doppler and growth scans (Royal College of Obstetricians and Gynaecologists, 2013). Such Doppler images are shown in Figure 6.

Umbilical artery Doppler has an important role in monitoring the foetal wellbeing in a high risk foetus with intra-uterine growth retardation. The umbilical artery Doppler serves both as a prognostic as well as a diagnostic tool, with an abnormal waveform (absent-end diastolic flow or reversed-end diastolic flow) predicting foetal compromise (Mone et al., 2015). Bekedam, Visser, & van der Zee (1990) looked at 29 foetuses with intrauterine growth retardation, and assessed the umbilical artery Doppler pulsatility index. Abnormal indices preceded late decelerations in 27 of the foetuses, and the median interval between an

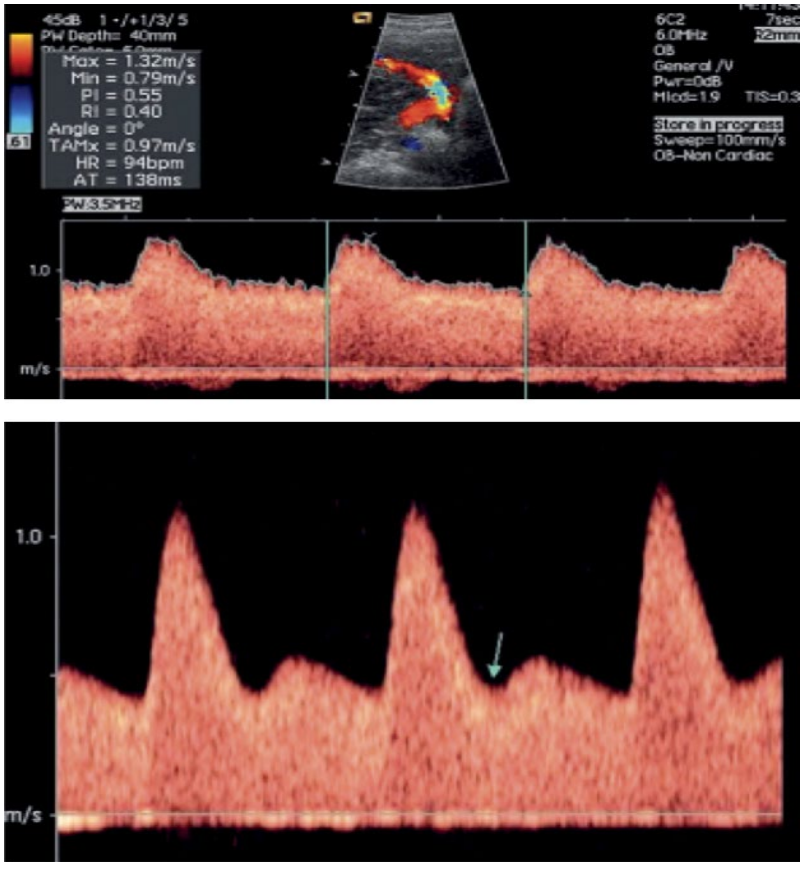


Figure 6. Second trimester uterine artery Doppler

1st image showing normal Doppler, 2nd image showing abnormal 'notching' shown by arrow

abnormal pulsatility index and a late deceleration in the foetal heart rate was 17days. Those foetuses with absent end diastolic flow were more severely growth retarded and had a worse perinatal morbidity and mortality rate. The role of abnormalities of Doppler flow has been extensively linked to pre-eclampsia and growth retardation conditions.

The role of ultrasound in obstetric triage is well established. In both the investigation of IUGR and pre-eclampsia , the role of additional predictors is ongoing (Mistry et al., 2013; Mone et al., 2015; Odibo et al., 2011; Royal College of Obstetricians and Gynaecologists, 2013). The end result is to be able to predict and triage for these conditions as early as possible so as to be able to improve the pregnancy outcomes. Thermography and temperature analysis in pregnancy may provide an additional parameter which may improve the outcome prediction models already in place with regards to triaging of these pathologies.

1.1.5 Thermography

1.1.5.1 Development of Devices for Temperature measurement in Humans

Temperature measurement devices have undergone extensive advancement over time, starting from the time of Galileo all the way to 1986, when Carl Wunderlich developed the clinical thermometer (E. F. Ring & Jones, 2006).

There has been a consistent movement away from the conventional glass thermometers towards using more disposable systems which can be used during routine clinical use. The use of liquid crystal sensors became available in the 1960's and gained popularity because of its re-usable and inexpensive nature.

In the early 1940's, the first electronic sensor for infrared radiation was developed. The technology was based on Sir William Herschel's discovery of infrared radiation, as well as his sons' work. John Herschel's experiments successfully created an image using solar radiation which he called a 'thermogram' – a term which is used today in relation to images made by thermal radiation. The leap for thermography technology happened when digital

computer processing solved the computing problems in archiving images in digital form enabling the selection of regions of interest and measuring temperature from the images obtained. It was after this that thermography was explored as a new medical temperature measurement technique (EFJ. Ring, 2006).

1.1.5.2 What is Thermography?

In thermography, naturally emitted infrared (IR) radiation from the sample object is measured using a 'thermal imager'. This is a sensor that converts infrared electromagnetic energy into an electrical voltage, which then is amplified and portrayed as a thermal image using the adequate computer program (Abreu de Souza, Gamba, & Pedrini, 2018). A major advantage of this method, as mentioned previously, is that sensing is remote and no contact with the subject is required.

It is now relatively easy to obtain an image with infrared thermography. In order to operate the infrared camera properly, to propose sound hypotheses and be able to interpret the thermal information appropriately, a basic understanding of the underlying physics is needed. Specifically of importance is the theory that explains how the practical translation of radiated energy to temperature is achieved where notions of blackbody radiation and emissivity become relevant (Priego Quesada, Palmer, & de Anda, 2017). The aim of what follows in this section is to highlight the physics behind these phenomena.

1.1.5.3 Electro-Magnetic Spectrum

Classically, electromagnetic radiation (EM) is the phenomenon of energy flow through space or a medium which can be described as the transmission of the oscillations of electric and magnetic fields set at right angles to each other. Apart from velocity which is a standard 2.998×10^8 ms^{-1}, the two main characteristics of electromagnetic radiation is the amplitude of the EM field and the frequency of the wave (Priego Quesada et al., 2017).

A spectrum of electromagnetic radiation generated by various sources of energy has been described. It is split up into a number of ranges or bands of wavelengths. Electromagnetic radiation known as infrared radiation or thermal radiation, is emitted by all objects with temperature above absolute zero ,within a range of 0.75–1000µm of wavelength (Lahiri, Bagavathiappan, Jayakumar, & Philip, 2012).

For the purposes of thermography, the infrared region is demarcated as approximate wavelengths of 0.8 µm to 1000 µm, lying between the 'red' edge of visible light up to below microwave frequency. Much of this range of the EM spectrum is in fact not suitable for infrared thermography, because its transmission is impeded by atmospheric gases (Usamentiaga et al., 2014). Indeed, for Infrared imaging, only a limited span of the infrared spectrum is made use of as presented in Figure 7. Three ranges of the IR spectrum are typically delineated for thermography. These are the shortwave (SW) region extending from

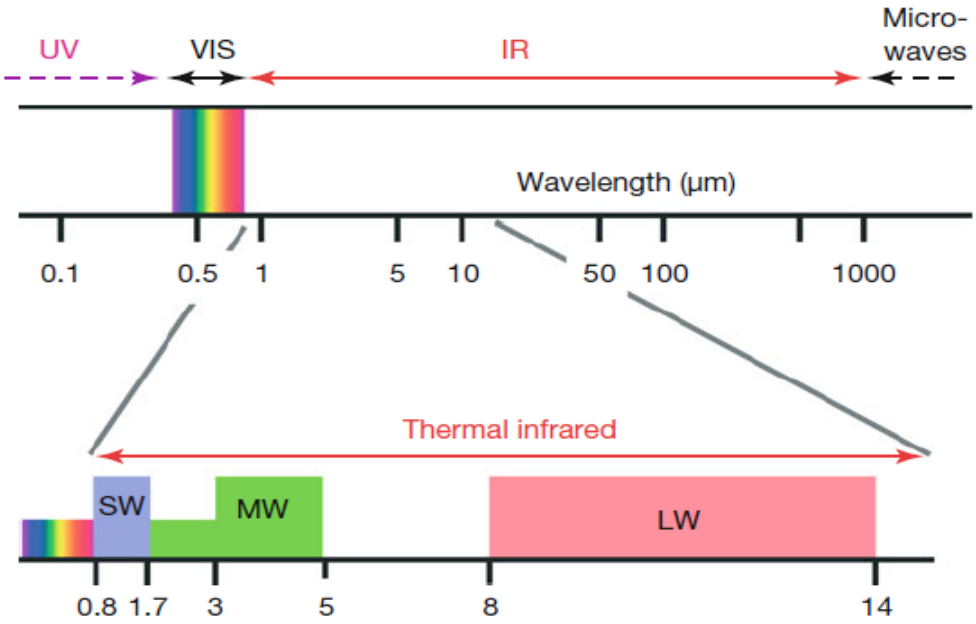


Figure 7. Expanded view of the thermal IR spectrum

(Vollmer & Möllmann, 2018)

wavelengths of 0.9 to 1.7μm, the midwave (MW) region around 3 to 5μm, and the longwave (LW) region around 8 to 14μm (Vollmer & Möllmann, 2018).

1.1.5.4 Physics of Thermography

When studying the temperature emitted by a human subject, there are various factors that are characteristic to both the human body as well as the environment of study. Skin temperature which is picked up by the thermal imager, results from the properties of the human skin which determine its emissivity and reflectivity. The form of energy emitted is infrared radiation which is directly related to the absolute temperature of the body surface itself - higher radiation intensity released as the temperature increases (Usamentiaga et al., 2014). The physics behind this phenomenon is explained below.

1.1.5.5 Thermal Radiation

Infrared energy that reaches a body mass could be absorbed, reflected or be transmitted across it, depending on the body's material properties (Figure 8). These features determine how the incidence of specific wavelength radiation per unit surface area of a body is

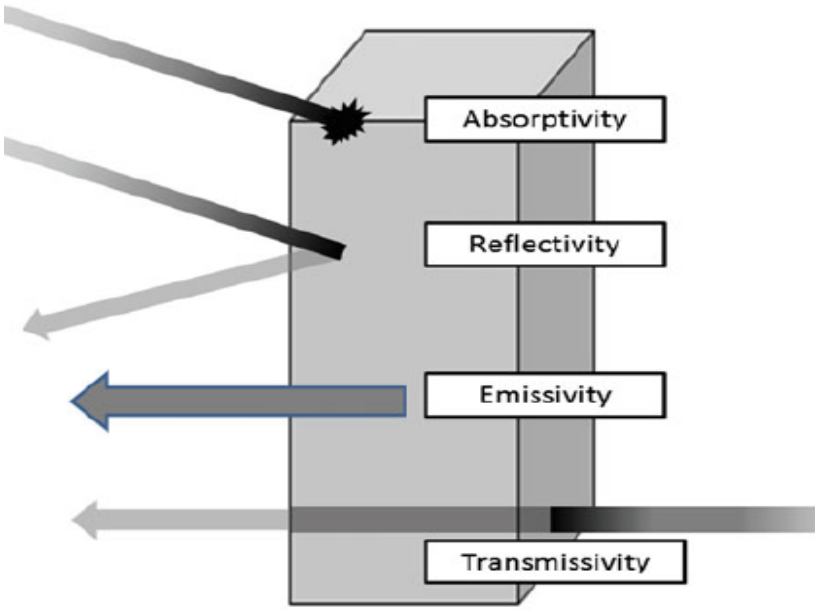


Figure 8. Radiative characteristics

(Priego Quesada et al., 2017)

dissipated (Priego Quesada et al., 2017). The total radiant energy is the sum of three fractions each of which is linked to a specific mode of dissipation namely absorptivity, transmissivity and reflectivity of the body. Absorptivity (α_λ) is the portion of the total irradiation that is absorbed by the body; reflectivity (ρ_λ) is the fraction reflected by the object; and transmissivity (τ_λ) is the portion of irradiation the body allows to be transmitted through it. In the event that there is no transmission through a medium ($\tau_\lambda = 0$) then it is said to be opaque (Priego Quesada et al., 2017).

These three properties are spectral dependent and considering the law of conservation of energy it follows that the sum of these three fractions at any wavelength is $\alpha_\lambda + \rho_\lambda + \tau_\lambda = 1$ (Usamentiaga et al., 2014).

When a body is opaque then this equation can be simplified to $\alpha_\lambda + \rho_\lambda = 1$, indicating that the irradiant energy is completely either absorbed or reflected. Looking at this from a radiation source view it can be put that the opaque body reflects the energy that it does not absorb and the equation written as $\alpha_\lambda = 1 - \rho_\lambda$ (Usamentiaga et al., 2014).

1.1.5.5.1 Blackbody Radiation

In his work 'The theory of heat radiation', Max Plank (1914) was the first to described the theory of radiant heat which is the essential foundation in the computation of temperatures on the basis of the spectral analysis of surface radiation (C. B. Pereira et al., 2018). The theory is based on the notion of a blackbody - a theoretical, perfect radiator of thermal energy emitting the maximum radiant power. Specifically, blackbodies are idealized surfaces that have the property of absorbing all incident radiation, independent of wavelength and direction. Additionally, at any specified temperature and wavelength, blackbodies emit more energy than any other surface. Finally, blackbody radiation is a function of wavelength and temperature however it is independent of direction (Bergman & Lavine, 2017). Blackbodies being ideal absorbers and emitters can be used as standards in radiometry

(Vollmer & Möllmann, 2018). Objects which have null transmissivity and reflectivity ($\tau_\lambda = 0$, $\rho_\lambda = 0$) are therefore blackbodies where all incident radiant energy is absorbed ($\alpha_\lambda = 1$) (Usamentiaga et al., 2014).

The calculation of blackbody emissive power of thermal radiation across the spectrum is possible on the basis of these assumptions. This function known as the Plank distribution, was first determined by Max Plank (Plank, 1914) and follows the equation:

$$E_{\lambda b}(\lambda, T) = \frac{2\pi hc^2}{\lambda^5 \left(e^{\frac{hc}{\lambda kT}} - 1 \right)}$$

Here, $E_{\lambda b}$ denotes the emissive power of the blackbody per unit area per unit wavelength ($Wm^{-2}\ \mu m^{-1}$), λ is the wavelength (μm), T stands for the surface temperature (K), $h = 6.626 \times 10^{-34}$ Js and $k = 1.3807 \times 10^{-23}$ JK^{-1} are the Planck and Boltzmann constants and $c = 2.998 \times 10^{8}$ ms^{-1} is the speed of light in vacuum. $E_{\lambda b}$ is a function of λ, which means that the total emitted radiation is distributed continuously over wavelength. At any wavelength the magnitude of the energy emitted is contingent on the blackbody temperature T and increases with increasing temperature (C. B. Pereira et al., 2018).

The spread of blackbody emitted radiation at discrete temperatures is shown in Figure 9. The graphs indicate per wavelength and temperature the amount of energy that is radiated. At any wavelength the magnitude of the energy depends on the blackbody temperature T and increases with increasing temperature. Furthermore, there is an counter relationship between temperature and the wavelength at the apex of the distribution with relatively more emission power evident at shorter wavelengths as the temperature rises (Usamentiaga et al., 2014).

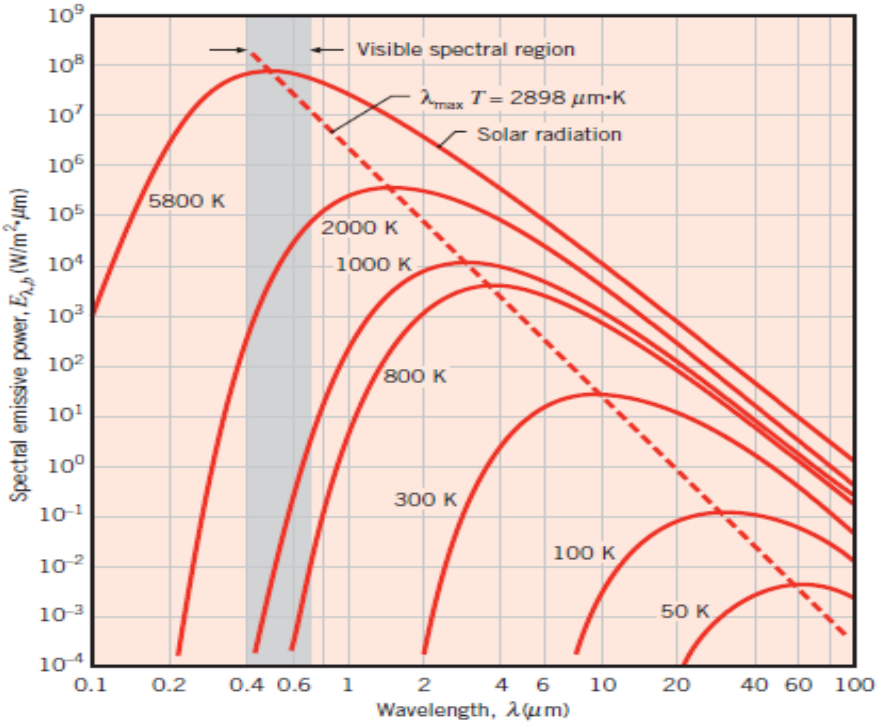


Figure 9. Spectral blackbody emissive power

(Bergman & Lavine, 2017)

Wavelength λ_{max} corresponding to the spectral distribution peak is dependent on temperature and can be calculated by differentiating the Plank equation ($\frac{dE_{\lambda b}(T)}{d\lambda} = 0$) which locates the maximum. Doing so leads to Wien's displacement law where:

$$\lambda_{max} \times T = 2897.8\ \mu m\ K$$

The dashed red line in Figure 9 joins the loci of points described by this law and shows that the maximum spectral emissive power is shifted towards shorter wavelengths as the temperature rises (Bergman & Lavine, 2017).

Planck's law describes the ultimate black body, and integrating this for all frequencies results in Stefan-Boltzmann's law. This states that the total energy E_0 emitted per unit surface

area of a black body per unit time is directly proportional to the fourth power of the black body thermodynamic temperature T

$$E_b = \sigma T^4$$

where $\sigma = 5.67 \times 10^{-8} \mathrm{Wm}^{-2}\,\mathrm{K}^{-4}$ is the Stefan-Boltzmann constant. On the basis of this physical law, the temperature of a body mass surface could be readily determined based on its measured emitted power per surface area. Broadly, sensors used in thermographic cameras operate on this principle (C. B. Pereira et al., 2018).

1.1.5.5.2 Emissivity & Reflectivity

A blackbody is an ideal theoretical concept – in the real world no physical object at a particular given temperature can emanate the full thermal radiation possible at that temperature. However, this actual emission of heat radiation off a real object can be calculated by applying a factor to the theoretical blackbody radiation that represents the effect on radiation of the object material. This factor is called the emissivity (ε) of that material. The emissivity of an object can be defined as the proportion of the measured quantity of radiation given off from the body to the theoretical emission from a blackbody at the same temperature (Vollmer & Möllmann, 2018).

Mathematically the emissivity of a material can be described formally for any given wavelength λ , as the actual body radiant energy divided by the theoretical blackbody radiation at the same temperature and can be represented as

$$\epsilon_\lambda = \frac{E_\lambda}{E_{\lambda b}}$$

As seen earlier, all of the energy striking an opaque body is either absorbed or reflected - consequently $\alpha_\lambda = 1 - \rho_\lambda$. If the absorption value is less than 1, which is the value of α for blackbodies, the material object is denoted as a grey body (C. B. Pereira et al., 2018).

Assuming further that emissivity is fixed and independent of wavelength, then the Stefan–Boltzmann formula for grey body radiators resolves to

$$E = \epsilon\sigma T^4$$ (Usamentiaga et al., 2014).

While real objects cannot be considered grey bodies, it can however be assumed that for small wavelength ranges, their emissivity is constant. Within this limitation, real objects can be treated as grey bodies. Thus, by taking their mean emissivity across short spectral spans – in practice the ranges where infrared sensors operate – and because for solid materials emissivity is a slow-varying function of λ, the Stefan–Boltzmann formula for real body radiators can be assumed to be the same as for a grey body (Usamentiaga et al., 2014). The comparative emissivity of a blackbody, a grey body and a real body is shown in Figure 10.

Although thermal imaging cameras allow inputting the emissivity of the body under measurement, this value is often not readily known (C. B. Pereira et al., 2018). One way of estimating a body's emissivity is by using Kirchhoff's law which states that in general, at any

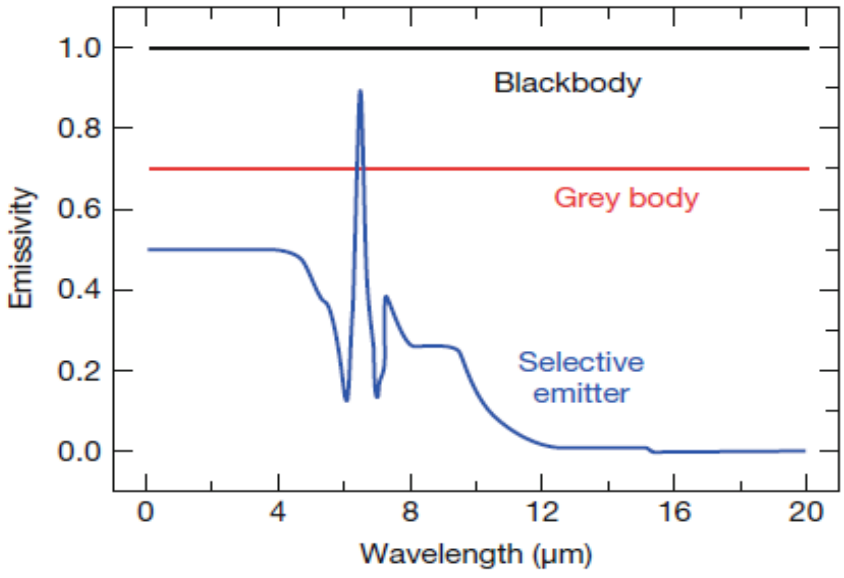


Figure 10. Comparative spectral hemispheric emissivities

(Vollmer & Möllmann, 2018)

particular temperature and wavelength a material's emissivity equals its absorptivity and could be stated as

$$\epsilon_\lambda = \alpha_\lambda$$

As stated in section 1.1.5.5 above, the law of conservation of energy requires that the sum of absorptivity (α_λ), reflectivity (ρ_λ) and transmissivity (τ_λ) as fractions of the incident radiation at any wavelength is

$$\alpha_\lambda + \rho_\lambda + \tau_\lambda = 1$$

For opaque materials and isothermal conditions then substituting for α, it follows that

$$\varepsilon = 1 - R$$

In other words, the emissivity can be estimated directly from known total reflectivity values (Vollmer & Möllmann, 2018).

1.1.5.5.3 Human Body Emissivity

Diverse studies (Steketee, 1973; Jones, 1998) have been carried out to determine the emissivity of the human skin tissue which indicate a consistent value of 0.98 ± 0.01. This value was determined for black skin, white skin and burnt skin, and no significant difference was found between them. The human body emits thermal radiation from mid-infrared to the microwave band with maximum emissive intensity at a λ ranging from 9μm to 10μm (Kurt Ammer, 2008). In fact the studies show that the human skin can be considered as a grey body with an emissivity close to that of a perfect black body and the emissivity coefficient of 0.98 can be applied in the infrared spectral range (Skouroliakou, Kalatzis, Kalyvas, & Grivas, 2017).

In medical applications where the surface under measurement is the human skin, blackbody theory can be used in quantifying temperatures (C. B. Pereira et al., 2018). However, internal organs like the heart have an emissivity which is markedly lower than

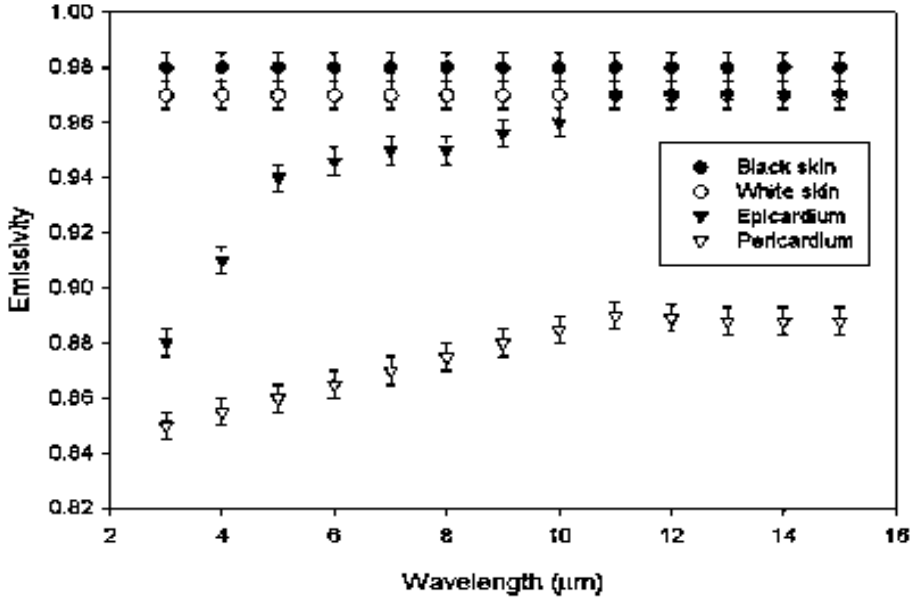


Figure 11. Emissivity of human skin

(Jones, 1998)

unity particularly for mid-wave λ's as shown in Figure 11. This is a salient consideration when thermal imaging is utilised for surgical purposes (Jones, 1998).

1.1.5.5.4 Temperature Measurement

The Stefan-Boltzmann law for blackbody radiation stated above ($E = \epsilon\sigma T^4$) applies for a body in vacuum and is the main relationship that allows computation of thermographic temperature measurement. However, for practical applications where an accurate temperature record is required, consideration of salient factors affecting assumptions need to be reviewed. A thermographic camera sensor detects all radiation reaching it and not just that from the body under study. Consequently, the recorded energy is made up of the object emission and in addition the reflected emission from environment and atmospheric sources and emission transmittance through the atmosphere. This indicates that the Stefan-Boltzmann law can be generalized for grey bodies in air as

$$E_{measured} = \epsilon_{obj}\tau_{atm}\sigma T_{obj}^4 + (1 - \epsilon_{obj})\tau_{atm}\sigma T_{env}^4 + (1 - \tau_{atm})\sigma T_{atm}^4$$

(C. B. Pereira et al., 2018), where the suffixes *obj*, *atm* and *env* refer to the object, the air and the environment respectively.

The transmittance of air τ_{atm} is commonly valued factoring the distance of the objective from the camera and the relative humidity of the surroundings. This value is practically very near to unity for λ's of interest and so the factor $(1 - \tau_{atm})$ is close to zero. Consequently this parameter has insignificant effect on the temperature readings (Usamentiaga et al., 2014) and the measured energy can be simplified to:

$$E_{measured} \cong \epsilon_{obj}\sigma T_{obj}^4 + (1 - \epsilon_{obj})\sigma T_{env}^4$$

The effect on EM power attenuation by atmospheric transmission is shown in Figure

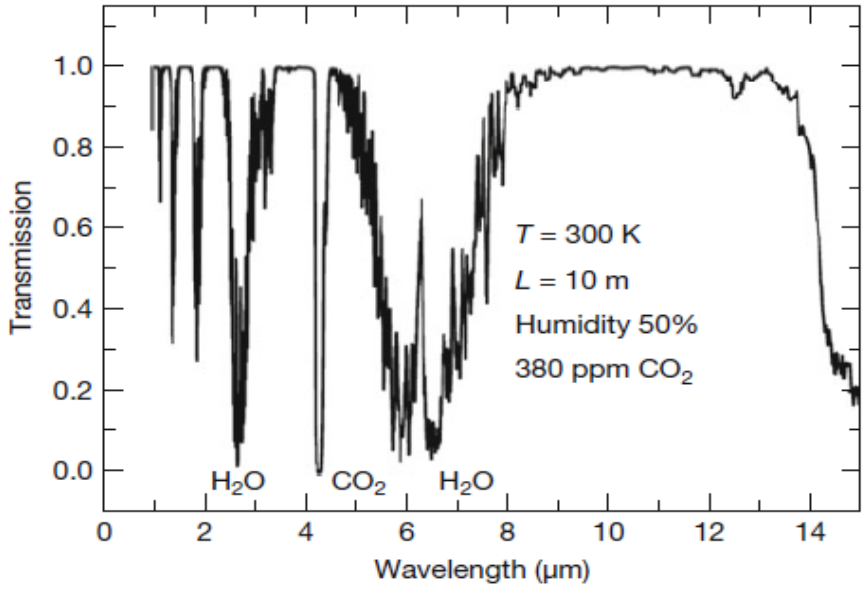


Figure 12. IR atmospheric transmission

(Vollmer & Möllmann, 2018)

12.

The factor of reflected infrared emission from the surrounding environment can be significant particularly when the reflectivity of the objective is high (Usamentiaga et al., 2014). Examples of thermal radiation sources of such emission are the sun, heating appliances or light bulbs. It is thus crucial when using thermographic equipment to ensure avoidance of such sources near the body being measured (C. B. Pereira et al., 2018). When

such safeguards have been taken rendering $(1 - \epsilon_{obj})$ to be close to zero, then the temperature of the objective can be calculated from the equation:

$$T_{obj} \cong \sqrt[4]{\frac{E_{measured}}{\sigma \epsilon_{obj}}}$$

Nonetheless this issue with reflectivity comes to play when the skin is covered by for example ultrasound gel or even water. Here the reflectivity is the property of the substance itself and not the skin, thus altering the temperature emitted and recorded by the thermographic imager (Ludwig, Formenti, Gargano, & Alberti, 2014) and appropriate caution needs to be exercised when setting up experiments and interpreting results. So if such cases of topical application of ointment, gel or solution on the skin cannot be precluded then correction of temperature readings are necessary because of the resulting alteration of emissivity. This is necessary because otherwise there is a high risk of getting biased measures of temperature, especially where the reflected environmental temperature is very different from that of the objective body (Bernard, Staffa, Mornstein, & Bourek, 2013).

Radiation emitted from a surface within a limited temperature range can be approximated to

$$E_{measured} \cong h_r \epsilon_{obj} (T_{obj} - T_{env})$$

by using a linearized radiation heat transfer coefficient h_r. For human skin thermal measurements in normal indoor environments and temperatures, h_r has been estimated at 4.7 W m^{-2} (Arens & Zhang, 2006).

1.1.5.6 Thermographic Imagers

E. F. J. Ring (2010) and E. F. Ring & Jones (2006) described in detail the history of thermographic imaging. They spoke about the start of thermographic display in the form of liquid crystal technology using Polaroid cameras. The technology was then advanced by the physicists Schwamm and Reeh, who developed the first recorded thermography system

which was used for medical purposes. In Great Britain, the first medical thermography system was developed by the Middlesex Hospital in London and the Royal National Hospital for Rheumatic disease in Bath. This was called the Pyroscan. It did have some pitfalls, mainly the poor image quality and the delay in recording an image. The research continued, and early cancer research was the driving force to developing newer technologies. With the development of computer processors, the image was continuously enhanced and the addition of the colour component improved the interpretation of the image. It was again in Bath where the system was digitised onto a colour screen. This digitisation allowed other facilities such as archiving of images, selection of regions of interest on the image, and also temperature measurements at particular points on the image.

1.1.5.7 Different Methods of Thermographic Imaging (static/dynamic)

There are various existing technologies in use for thermography use in healthcare. There were classified into four groups by Jiang (2005), which were the following: (1) low level processing (2) high level processing (3) human thermal modelling and (4) computer aided diagnoses.

Low level processing is described as techniques which processes a still thermographic image such as enhancing an image or measuring a point temperature on that image. High level processing can be divided into static methods or dynamic methods. In the case of static methods, a single image is taken; however, the interpretation depends on spacial distribution, meaning the symmetric distribution of temperature is taken into account by the processing programme. This method corrects for the temperature asymmetry of contralateral sides of the body by assuming that they are in fact symmetrical by comparing to normative reference values. It is pointed out by Jiang (2005) that many environmental and patient factors need to be taken into account when carrying out these studies and these will be discussed later on in the Methodology chapter. The dynamic method is able to incorporate the behavioural

stresses on the skin together with the spacial aspects already seen in the static method. A typical example of this is functional imaging where the subject is exposed to thermal stress such as hot or cold water, and the rate at which the temperature reaches equilibrium once again is measured (E. F. J. Ring, 1995). Dynamic area telethermometry is another method of high level processing where hundreds of consecutive IR images are quantitatively analysed and due to the amplitude of frequency of the components, neural effects can be separated from haemodynamic effects. This method has been applied to breast cancer studies for example, because it enables the detection of abnormal temperature of the skin whilst assessing spacial distribution of the skin perfusion, and this allows quantitative analysis of the organ studied.

1.1.5.8 Thermographic Cameras

In their contribution to the book "Topics in Medical Image Processing and Computational Vision", Vardasca & Simoes (2013) described the components of a thermal camera which are essential to improved equipment for medical use.

The main components of a thermal camera are:

- The optics. This is where the infrared energy emitted by the subject is focused on the detector. These are made of either silicon or germanium. The silicon optics detect shorter wavelengths between 3-5µm, whilst the germanium optics detect longer wavelengths between 7-14µm.
- The Infrared detector array. This is where the infrared energy is converted into an electrical signal and is the aspect of thermographic cameras where there has been most evolution throughout recent years. The first scanning model had only one sensor element which scanned the whole image column by column and line by line – a long and inefficient process. The most recent method developed was the Focal Plane Array (FPA) where there is a matrix of

sensor elements which record the whole image at once and produce faster and better quality images.

- The shutter system. This is where any image correction is carried out.
- The digital signal processing (DSP) unit. This is the electrical part of the camera which processes an electronic signal in order to produce the image and be able to perform temperature calculations.

There are two types of camera groups – cooled and uncooled. The cooled type, require cooling to function. They function by refrigeration using Stirling or thermoelectric coolers. Their main downside is their manufacturing and functioning costs. They can be very expensive and bulky, and have a limited operational life of 8 to 15 hours. They mainly operate within the shorted wavelength range of 3-5µm. The main advantage is that they do provide superior image quality and are much more sensitive to minor temperature changes of the environment being imaged.

Uncooled cameras on the other hand, function at ambient temperature or temperatures close to ambient temperatures. These cameras operate at longer wavelengths of 7-12µm and produce lower quality images compared to the cooled cameras. However, having smaller dimensions they are lighter, handier and simpler to use.

1.1.6 The use of Thermography in Medicine

Once thermography started to gain popularity in the 60's and 70's, there was a movement towards standardising the use of this modality in the field of medicine. The European Thermographic Association was formed in 1972 and held their first major conference in Amsterdam in 1974. It was during this conference that various study groups where formed and guidelines of practice where formulated (E. F. J. Ring, 2007). The Japanese Society of Biomedical Thermology was also founded in 1978, and the American Academy of Thermology in 1971, these institutions also having guidelines of practice and use

of thermography (E. F. J. Ring & Ammer, 2015). The starting of such institutions highlights the increasing importance of this modality in medicine. Thermography was and still is increasingly used in various fields in medicine.

1.1.6.1 Thermography and its use in Medical Specialities

One of the larger areas of study is in breast cancer detection. In 1982, the US food and drug administration (FDA), approved infrared thermography as an adjunct tool for diagnosis of breast cancer (Lahiri et al., 2012). A thermogram can be used in conjunction with mammography, which is currently the gold standard investigation. The latter will show the anatomy of the breast but a thermograph can pick up the physiology and areas of high metabolic activity. E. Y. K. Ng & Sudharsan (2004) demonstrated that the combination of the two modalities increased the sensitivity to 93% as compared to a mammogram alone which has a sensitivity of 83%. E. Y. K. Ng (2009) also showed that abnormal breast tissue can represent significant biological abnormalities, because of the increased blood supply and metabolic rate in the abnormal breast tissue. These areas can be visualised by IRT as 'hot spots' - Figure 13 is a typical visualization. 86% of cases of non-palpable breast cancers also showed areas of hyper vascularity and hyperthermia (Lahiri et al., 2012) .

Rheumatology, together with breast pathology, was one of the earliest medical fields

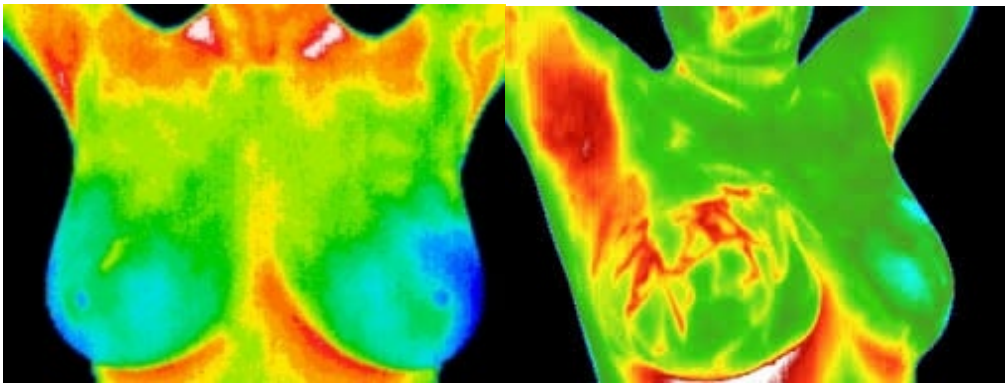


Figure 13. Breast Thermogram

Normal Breast Thermogram (left) vs abnormal breast thermogram showing angiogenesis (right).

that were investigated using thermography. Ring & Bacon (1974) carried out one of the

earlier studies looking at rheumatoid arthritis and compared the thermographic images of normal knees, acutely inflamed knees and those treated with a dose of steroid. They created a thermal index by using the isothermal images and this enabled them to observe the effect of treatment in a quantitative manner. Ring, Collins, Bacon, & Cosh (1974) used this same index to compare different anti-inflammatory treatments, mainly the comparison between oral and local injection. The action of paracetamol was also looked at and it was seen that the thermographic index continued to rise in the inflamed joint, showing that this method can distinguish between analgesia and anti-inflammatory action. More recently, Anderson, Moore, Lunt, & Herrick (2007), investigated the use of thermography to differentiate between primary and secondary Reynaud's phenomenon. By carrying out a retrospective analysis of clinical findings and thermographic images of 56 patients with primary Reynaud's phenomenon, 21 patients with undifferentiated connective tissue disorders and 45 patients with systemic sclerosis, they showed an 82% sensitivity and 82% specificity in identifying Reynaud's phenomenon secondary to systemic sclerosis, with a negative predictive value of 89% and a positive predictive value of 73%. These studies show that thermography can be used as an adjunct to already existing investigative devices in rheumatology, in order to improve the treatment monitoring and diagnosis of these conditions.

Diabetic foot is the major complication of diabetes. It is caused by peripheral neuropathy and peripheral vascular disease. Branemark (1967) looked at thermographic patterns of hands and feet in diabetic patients. All 16 patients showed a different thermographic pattern of the feet when compared to non-diabetic patients, especially showing reduced emissions over the toes and meta-tarsal regions. Bharara, Cobb, & Claremont (2006) reviewed the research carried out using different techniques of thermography to detect early signs of peripheral vascular disease due to diabetes. They discussed how thermography can be useful clinically because of the effect of progressive degeneration of sensory nerve pathways which affect both thermoreceptors and mechanoreceptors. A focus of research has

been the early risk detection of ulceration especially on the planter aspect of the foot. Armstrong (1997) looked at 143 subjects suffering from diabetes, dividing them into those with asymptomatic sensory neuropathy, neuropathic foot ulcers and neuropathic fractures. Over a period of 22 months, they evaluated the foot temperatures using a hand held infra-red temperature probe, with contralateral foot temperatures measured as controls. It was found that there was significance in the difference between the contralateral and affected foot in the neuropathic fracture and neuropathic ulcer group. There was no significant difference in in the group with asymptomatic neuropathy. The potential addition of the use of thermography as a tool to be used in clinical practice for prevention of ulcers, was tested by Avery et al. (2004) when they found that the addition of thermography to their standard therapy, had significantly less diabetic foot complications when compared to those who had standard therapy (2% vs 20%, $p = 0.01$).

In their extensive research review, Lahiri et al. (2012a) describe the investigation of the use of thermography in other areas such as dermatology, vascular surgery, plastic surgery and musculoskeletal medicine to name a few. This demonstrates that the interest in the how this device can be utilised to enhance investigation and treatment of medical conditions holds a constant interest in the medical community.

1.1.6.2 Protocols

The AAT (American Association of Thermography) has produced a detailed guideline for the operator of thermography when investigating breast pathology, including all the necessary assessments to carry out with the patient, as well as equipment requirements. These will be presented in further detail in the Methodology section.

1.1.6.3 The use of Thermography in Obstetrics

There has been very little published work when it comes to the use of thermography in human obstetrics. The early studies published focused on trying to localise the placenta by

using thermography. Millar (1966) published one of the initial studies, where they looked at the use of thermography as a method of localising the placenta in pregnant women. In their study, they scanned 150 patients using the Pyroscan thermography apparatus, and compared the images obtained with the location of the placenta on delivery. Out of the 150 patients, only 23 patients were delivered by caesarean section meaning that the placenta could be adequately localised and compared. The results showed that 16 out of those 23 were not localised on thermography. The study did however highlight some issues which were, and to some degree, remain, pitfalls in the use of thermography in pregnancy. Firstly, Millar acknowledged that the environment posed a potential difficulty in recording images, specifically in maintaining an adequate ambient temperature which would not disrupt the thermographic recording, as well as the time required for acclimatisation. The method used in this study to quickly cool down the abdominal temperature to reach stable temperatures was to apply a damp cloth over the abdomen for 10 minutes. Although this is not a method that is routinely used, it showed the importance of this process in obtaining an ideal environment for the study to take place. Another relevant point that was given importance in the methodology of this study was that the thermographic apparatus was positioned in such a way that the lower segment of the abdomen, and thus the uterus, would be adequately visualised. This demonstrated the difficulty in capturing the image of a gravid abdomen, when compared to a non-pregnant abdomen.

Liu & Blackwell (1979) carried out a similar study, but instead of using a thermographic camera, they explored the use of liquid crystals. In their study, they analysed 43 pregnant patients. In each of the patients, the abdomen was ‘divided’ into 7 segments and temperature readings using liquid crystal films were carried out. These temperature patterns were then compared with ultrasound imaging of the abdomen, in relation to the placental location. The premise of the study was that the placenta, being a structure that emits large amounts of heat, would demonstrate the greatest temperature change on the liquid crystal

films, when compared to areas where the placenta was not located. In this way, the area of placental location could be determined by temperature change within the segments of the abdomen. The results were promising in that 23 out of the 43 patients had good correlation with the placental location. Posteriorly located placentae were 'assumed' by exclusion when it came to temperature measurements. There were however some issues that were highlighted namely, the issue of obesity and that of gestation. It was noted how obesity was seen to be a difficulty in the measurement of temperature, since in these areas, the temperature changes from cold to warm was too rapid and resulted in a difficulty in discriminating between normal skin temperature and hotter areas due to placental location. Regarding gestation, it was seen that in earlier gestations, it was easier to note temperature change and diagnose placental location, most likely due to the larger placenta to uterine size ratio at that stage of pregnancy.

These studies did shed light on potential difficulties and issues that can be encountered when it came to thermography in pregnancy. Their main limitation however was the fact that they were carried out in the early years of thermography research and adequate protocols and methodologies were not yet well developed for studies such as these. There was however very little advancement in research in the field until the research carried out by Simoes, Vardasca, & Nogueira-Silva (2012). In this study, they looked at 41 pregnant women at gestations between 34 and 40 weeks. The patients were divided into groups according to age and BMI, and an acclimatisation time of 15 minutes was carried out before the study. Anterior and posterior thermographic images of the trunk and abdomen were taken using a thermographic camera. These images were then analysed by using the thermographic software. Six ROIs (Regions of Interest) where defined on the Anterior aspect and four ROI's were defined on the posterior aspect as shown in Figure 14.

The statistical analysis showed that there was strong repeatability in the ROI measurements with an ICC of > 0.9 for all the selected ROI's. There was however, no

correlation of temperature with age. In non-pregnant studies, temperature showed a correlation with ages over 60 (Niu et al., 2001) due to the change in blood flow to the skin with age. This is reflected in the results of this study. Another interesting finding was that there was no correlation with BMI. This was addressed by the authors as being due to the reduced area of the body that was studied and more research was required in this area.

This further research was carried out by Pereira, Nogueira-Silva, & Simoes (2016). In the study, they looked at 61 pregnant patients and mapped out 31 regions of interest (ROI) for the whole body as shown in Figure 15.

The patients were also divided in groups by age, gestation and BMI. Temperatures were recorded in the same way as was done in the previously described study. In this study, the reliability of the ROI readings was found to have an ICC of more than 0.9, remaining consistent with the previous study. This study did differ from the previous one regarding BMI and age. There was a moderate correlation of temperature with maternal age ($r = 0.325$; $p = 0.01$) and there was a low correlation of temperature with high BMI ($r = 0.184$; $p = 0.01$). The correlation with BMI did reflect the findings in the literature (Adam, 1989). The moderate correlation with maternal age was a new finding, when compared to the results of the previously described study. The latter could be attributed to the way the patients were grouped. As opposed to the previous study, where the groups were independent of other

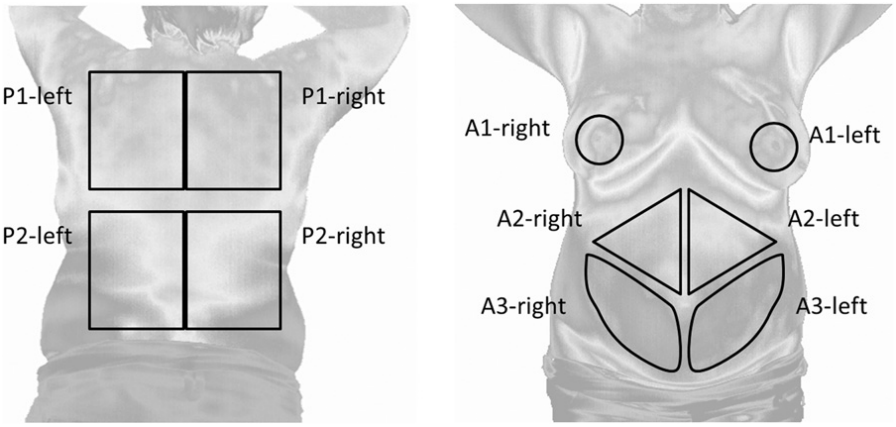


Figure 14. Regions of interest

Simoes, Vardasca, & Nogueira-Silva (2012)

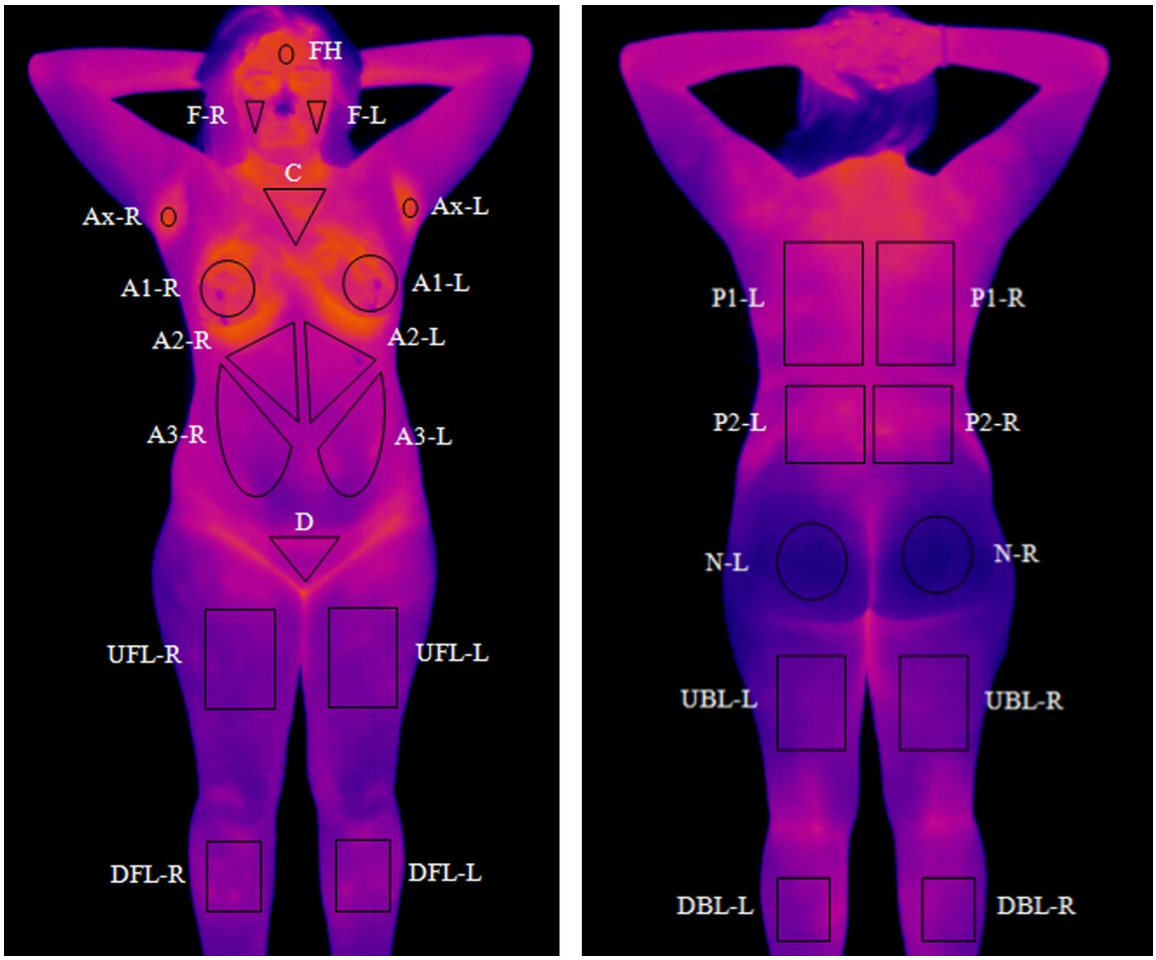


Figure 15. Anterior and Posterior ROI's

Pereira et al. (2016)

variables, in this study, the groups where not homogenous when it came to gestation and BMI, and these factors may have altered these results.

One important consistency throughout these two studies was the criteria set for the protocol of preparation for the patients. It was ensured that patients were not allowed to smoke, eat heavy meals, exercise or apply ointment to the body, on the day of the study.

The research carried out in pregnant humans with thermography to date, has shown much improvement both in the protocols used, as well as the inevitable advancement in software and cameras used. In the current study, the researcher wanted to build on these existing studies, and increase the knowledge on certain aspects of this field, in order to provide more information for future research in obstetrics and thermography.

1.2 Rationale for the Proposed Research

The aim of the research undertaken was to assess thermography as a modality in the setting of the use in obstetrics in humans. As discussed in the previous section, there has been a vast amount of work carried out to investigate the use of medical thermography, however in obstetrics the work has been minimal. The largest body of work was perhaps in the early days of thermographic research were the majority of focus was on placental localisation (Liu & Blackwell, 1979; Millar, 1966). During this time, the thermographic cameras were still in their early stages, as was the field of ultrasound in obstetrics, thus making these studies, although important, rather outdated. Looking at the more recent studies, it seems like thermography was being utilised to research specific obstetric conditions, rather than looking at thermography as a device in its own right for its use in obstetrics in healthy controls. For this reason, this research focussed on looking at the baseline temperatures in the pregnant patient, and how this would correlate with measurements on ultrasound, this being a well-established investigative device in the field of obstetrics.

The study was aimed at looking at a broader picture of how thermography can be interpreted in the pregnant human female. Early on in the research, it was identified that different aspects of pregnancy such as gestation, maternal age, maternal BMI, foetal position, placental position would potentially have a bearing on thermographic interpretation. It was also important to look into other basic factors such as timing of temperature acclimatisation in order to be able to create a standard protocol for thermographic studies in pregnancy.

Since the standard comparative device that was used in the experiment was ultrasound imaging, the primary pregnancy characteristic that was chosen to be observed in a direct comparison between the thermographic image and the ultrasound image, was the liquor pools around the foetus. This parameter was chosen in view of the standard method of measurement by ultrasound which entails ‘dividing’ the uterine cavity into four quadrants and measuring the liquor pools in each of the four quadrants, with the resulting summation of all

four being the Amniotic Fluid Index (AFI). This lent itself well to defining thermographic Regions of Interest (ROIs) when analysing the respective thermographic image. Other variables were also observed as will be described later on in the chapter.

The principal factors studied in this research, were the liquor pool depth in each quadrant, measured by ultrasound, and defined in centimetres and the temperature measurements on thermographic image at the respective quadrant, analysed by thermographic software, and defined in degrees centigrade. This was the case throughout all the phases of the research project.

1.3 Research Questions

The research questions set out to be answered by this study were:

- As the general exploratory question, does a relationship exist between the thermographic image data and the ultrasound image inferred information?

Specifically, the questions of interest are:

- Is there a correlation between the thermographic temperature level and the maximum depth of the liquor pool measured with ultrasound at the regions of interest (ROI's)?
- Do maternal and biological parameters influence the correlations and how do environmental parameters such as acclimatisation period affect this relationship?
- Is the relationship between thermographic temperature level and ultrasound measurement of liquor volume at ROI also present in pregnant patients suffering from diabetes? Do maternal and biological factors influence these correlations and how does an acclimitisation period affect this relationship?

In the next chapter, the research questions will be broken down according to each phase of the study, where specific questions are analysed. The approach to the research design and the methodology carried out for each part of the study will also be discussed.

2 Methodology

2.1 Overview

In the previous chapter, the topics of research were discussed with reference to the literature, followed by a broad description of the overall research question and objectives for the study undertaken by the researcher. This chapter gives an outline of the methodology and the approach to that same methodology used for the research study. Firstly, certain aspects of thermography research which need to be understood in relation to the study design are discussed. The chapter then moves on to discuss the various phases of the study and breaks these down into the more specific objectives and research questions, ethical considerations, the recruitment of participants, the instruments and setting used during the study. Also discussed are the procedures and protocols and the data and image analysis carried out for each phase.

2.2 Approach towards Research Design

In this part of the chapter, the salient factors that need to be considered when carrying out thermography research in humans are discussed. Each factor is further considered when describing the methodology of the individual phases of the research study.

2.2.1 Factors that influence IR Thermography use

Although the literature is rich in studies describing the use of IRT in human pathology, there is distinctively less research carried out looking at the healthy human cohort. There is even less data regarding the healthy pregnant human cohort, and this is where the focus of this study lies.

It is important to realise that working with IRT requires various factors which need to be accounted for during the use of the device, and each of these factors can influence the data collection and interpretation during research or clinical use. Fernández-Cuevas, Bouzas Marins, et al. (2015) divided these factors into three groups – environmental, individual and technical.

2.2.1.1 Environmental Factors

In medical IRT research, the temperature differences that are being measured are a factor of a degree. It is for this reason that environmental variables are required to remain as stable as possible across the experiment or clinical use. These environmental elements are amenable to some degree of control, unlike patient factors. The following are certain aspects of the surrounding environment in which the study is taking place, which should be controlled as much as is possible.

2.2.1.1.1 Room Size

The room choice must be such that this remains as neutral a space as is possible. The room is advised to be at least 2x3m in size so as to allow for the patient, researcher and equipment to fit comfortably (E. Y K Ng, 2009). The space should be sufficient for a patient of any size to be able to comfortably undergo testing with free range of movement, with the patient spaced equidistant from each wall during testing (International Academy of Clinical Thermology, 2015). It is suggested that rooms with large ceilings are inadequate in that they will then result in difficulty in maintaining a homogenous ambient temperature as part of the standardisation protocol (Fernández-Cuevas et al., 2015). It is also important to survey the room for any sources of infrared which may interfere in the thermographic recording. These include windows allowing external light into the room – these should be adequately covered with blinds to shut out as much light as possible; as well as incandescent light – these should be turned away from the patient or switched off and standard fluorescent lighting is preferred (International Academy of Clinical Thermology, 2015).

Following the above findings, the size of room used for the experiment was measured and arrangements were made so that for each phase of the study the same room is used for all the trials to minimize environmental confounding factors.

2.2.1.1.2 Ambient Temperature

The temperature of the room is required to be maintained as stable as possible, with a variability of temperature of ± 1˚C. The stability of the temperature is important in order to eliminate the possibility of shivering and sweating, which could both be sources of destabilisation in body temperature. It is suggested that the optimum temperature range should be kept between 18 and 23 degrees Celsius (International Academy of Clinical Thermology, 2015). There have been studies that showed that warmer temperature ranges, specifically between 22˚C and 24˚C, are recommended for studies involving peripheral regions of the body, whilst cooler temperatures are suggested in studies looking at inflammatory conditions (Fernández-Cuevas et al., 2015).

The ambient temperature is also potentially altered by the air conditioning vents within the room selected for the study. It is recommended that there are no direct draughts onto the patients and the ducting should be such within the room so as to distribute the air evenly over the room (E. F. J. Ring & Ammer, 2015). The various elements within the experiment, including the equipment used as well as the personnel involved, will all emit some degree of heat and this can potentially alter the room temperature. For this reason, the ventilation is important in order to provide a stable temperature which will not be too cold so as to avoid shivering and not too warm to result in perspiration (E. Y K Ng, 2009).

In the current study the room temperature was controlled by means of air-conditioning and kept at a mean of 22°C with no direct venting in the patient vicinity.

2.2.1.1.3 Patient Acclimatization

One aspect of the process of thermographic studies that is important not to be overlooked is the time it will take for the patient to acclimatise to the surrounding ambient temperature. This has varied immensely across different studies. Lahiri et al. (2012a) showed how the acclimatisation times varied between 5 minutes in some studies, to up to 30 minutes

in others. It is thus safe to say, that there is no consensus when it comes to the time required for acclimatisation, however, the various studies did highlight the thermal asymmetries between left and right sides of the body. Uematsu (1985) carried out one of the earliest studies comparing the contra-lateral sides of different parts of the body. A total of 64 contra lateral segments of the body were studied – 32 parts of the body. The difference in temperature was compared and found to be different in the different parts of the body, for example 0.12˚C on the forehead, and 0.23˚C on the knee (patellar aspect). It was also shown that there was more fluctuation in the peripheral structures, but overall the temperature remained stable between contralateral segments in healthy individuals, meaning that acclimatisation will not vary between symmetrical areas of the body. A more recent study by Ricardo Vardasca, Ring, Plassmann & Jone (2016), confirmed values of temperature differences between contralateral segments in the body which were similar to previous studies. The improved smaller difference in temperature difference was put down to regional as opposed to full body imaging, better patient preparation when compared to the older studies, more rigorous environmental preparation as well as the imaging equipment itself. It is also noted that a 15 minute acclimatisation time was used in this study.

One of the few studies (T. Pereira et al., 2016) that looked at the body symmetry in pregnant patients showed that there was a higher overall mean temperature in the upper regions of the body, namely the breast and axillary region. There was confirmation of similar temperature differences between symmetric regions in the body as were found in non-pregnant studies. The acclimatisation time here was also 15 minutes, however it must be noted that patients of various gestation age where recruited in this study.

These varying research outcomes result in an ambiguously wide range of acclimatisation time data in healthy pregnant women at a fixed gestation range. In view of this it was decided that an arbitrary but reasonable 60-minute acclimatisation period would be

used in this study in order to examine specific changes throughout this time, to observe the stabilisation patterns and to record when these are reached within the 60 minutes.

2.2.1.2 Individual Factors

The individual factors can be broadly divided into intrinsic and extrinsic factors (Fernández-Cuevas et al., 2015). In a standardised protocol, the extrinsic factors can be controlled by clear instructions to the patient and proper accountability within the study data once it is analysed. Intrinsic factors are more specific to the patients and cannot be controlled or eliminated for the purpose of the study itself. It is therefore important to note the possible effects it may have on the thermographic imaging results.

2.2.1.2.1 Intrinsic Factors

These are the basic characteristics of the subjects themselves. They range from biological to anatomical parameters and their effect on the thermographic studies is seen throughout the literature.

It has been shown that there is a difference in skin temperature between genders. Chudecka & Lubkowska (2015) looked at thermal maps for women and men and found that women have a significantly higher upper body skin temperature. This is also negatively correlated to BMI and percentage body fat. This correlation was true both in men and women, in several areas including abdomen, hand and thigh. In this study, the subjects are obviously female making this factor difference irrelevant.

Age has also been a factor that has extensively been shown to have a strong relationship with skin temperature. Most studies however tend to look at the temperature differences between extremes of age, such as in the case of Niu et al. (2001). In this study, it was shown that the average skin temperature of the elderly patients was slightly lower than that of younger subjects, with distal parts of the extremities having the largest difference. This is not relevant to this study since the subjects are younger patients and relatively similar

in age. In another study (Zaproudina, 2012), it was shown that age was not a factor that impacted the study significantly. The age of the subjects in this work ranged from 18 to 28 years, which is a closer range of age groups that was seen in our study.

Another intrinsic characteristic that has been shown to affect the skin temperature is the subject body mass. Savastano et al. (2009) looked at the thermal pattern characteristics of obese patients (taken as a BMI of > 30 kg/m^2) and non-obese patients. It was found that although there was no significant difference regarding core temperature, there was a significantly higher temperature measurement at the nail bed with obese patients. There was however, a significantly lower temperature measurement in obese patients for the abdominal skin. It was proposed that the higher subcutaneous adipose tissue in the obese group provides a significant insulation layer which then reduces the abdominal heat transfer to the surrounding environment. The increased heat release from the peripheries may aid in maintaining normothermia in these obese patients (Savastano et al., 2009). In pregnancy, it is known that a woman undergoes a degree of weight gain. There are various guidelines dictating the recommended weight gain and these were summarised and reviewed in a recent discussion paper by Siega-Riz et al. (2009). Based on the Institute of Medicine, the recommended increase in weight is relative to the initial weight prior to pregnancy as seen in Table 1.

Pre Pregnancy BMI Category	*Total Weight Gain (kg)*	*Rate of Weight Gain 2nd and 3rd Trimester (kg/week)*
Underweight (<18.5 kg/m^2)	12.5 – 18	0.51 (0.44 – 0.58)
Normal Weight (18.5 -24.9 kg/m^2)	11.5 – 16	0.42 (0.35 – 0.50)
Overweight (25 – 29.9 kg/m^2)	7 – 11.5	0.28 (0.23 – 0.33)
Obesity (≥ 30 kg/m^2)	5 - 9	0.22 (0.17 – 0.27)

Table 1. Gestational Weight Recommendations

The participants in the current study were mostly in the 2nd trimester and so weight gain was expected. It was therefore also expected that BMI and weight would play a part in affecting the skin temperature on thermographic imaging.

Finally, skin emissivity is an important intrinsic factor which affects the skin temperature detected by thermography. It is an important factor because it is a value required to be inputted into the thermography software, and therefore a standard value is needed. Many studies have been carried out to determine the emissivity of the human skin tissue (Steketee, 1973; Jones, 1998; Ammer, 2008; Skouroliakou, Kalatzis, Kalyvas, & Grivas, 2017). As discussed in the literature review section, the research shows that the human skin can be considered as a grey body with an emissivity close to that of a perfect black body and the emissivity coefficient of 0.98 can be applied in the infrared spectral range (Skouroliakou et al., 2017). The value was evaluated for white skin, black skin and burnt skin and no difference was seen between these different skin types regarding emissivity (Steketee, 1973). For our study, the emissivity was therefore set at a standard of 0.98. It was also decided that the skin type of each subject will be recorded by using the Fitzpatrick classification system (Fitzpatrick, 1975; Fitzpatrick, 1988) in order to assess the skin temperature in relation to this.

2.2.1.2.2 Extrinsic Factors

These are factors that affect the skin temperature for a period of time and can be altered by being included or excluded from the protocol accordingly. Such factors that affect the skin temperature due to ingestion which are relevant to our study are medications, smoking and alcohol.

There is no simple list of medications to avoid during thermographic testing. Many studies actually assess the effect of various clinical states with or without treatment, be it analgesics, anti-inflammatories, vasoactive treatments or hormonal treatments. One of the

initial studies that looked at the effect of inflammation and anti-inflammatory treatment in rheumatoid arthritis by using thermographic imaging (Collins et al., 1974), clearly demonstrated that corticosteroid treatment resulted in a different temperature pattern in rheumatoid affected joints, thus showing that temperature can be used as an objective marker of treatment. Giani, Rocchio, & Tavoni (1989) demonstrated that thermography could be used as an effective tool in evaluating the use of NSAIDs in musculoskeletal injuries. Vasoactive treatments such as aspirin and clopidogrel were also shown to alter core temperature, as seen in the study carried out by Bruning, Dahmus, Kenney, & Alexander (2013). They showed that the use of these medications versus placebo resulted in a shift of onset of peripheral thermo-effector mechanisms towards higher body temperatures during exercise. Pregnant patients may sometimes be on such treatment; however, as part of the exclusion criteria of the main study, the patients were chosen so as to not have any medical conditions or be on any such treatment. During Phase 3 however, the patients, being diabetic, may be on treatment for this condition and the study will evaluate in part the effect of such treatment on the temperature readings.

Smoking is another factor that must be included in the assessment of the subject during the study. It is well known that nicotine has a vasoconstrictive action on the skin and this results in a reduction in temperature. It also increases the pulse and blood pressure of the smoker (Fernández-Cuevas et al., 2015). The effect of smoking on the thermographic imaging of the hands over time by Gershon-Cohen & Haberman, (1968) which showed a temperature drop of 1˚C eight minutes after smoking a cigarette, with a persistent drop in temperature of up to 3˚C thirty five minutes after. On the other hand, another study did not find any significant difference between smokers and non-smokers in their cohort. They did however attribute this to the small number of smokers in their sample group (Christensen, Vaeth, & Wenzel, 2012). In the research presently undertaken, the patients were asked about

their smoking habits and this was documented and analysed to assess their effect on the temperature readings obtained on thermography.

Alcohol consumption also has been associated with increased vasodilation of skin blood vessels, and a resultant increased skin blood flow. Ewing, Davison, & Fergason (1973) demonstrated this using infrared thermography, showing that subjects after alcohol consumption exhibited an increased temperature and a more 'diffuse' thermographic image when compared to normal. Furthermore, a recent study showed that the thermal response to alcohol intake differs between different areas of the body, with the hands having a maximal increase of 1˚C in temperature after 15 minutes of intake, whilst the temperature of the knees increased by 0.3˚C (Melnizky, P., & Ammer, 2000). In view of the effect alcohol intake has on the skin temperature, alcohol consumption must be included in the questionnaire protocol or in the preparation protocol when designing any study.

EFFECT OF DIABETIC TREATMENT

Other extrinsic factors that may affect body skin temperature include the application of ointments and creams. As discussed in a previous section, this is an issue with reflectivity and comes into play when the skin is covered by for example ultrasound gel or even water. This renders reflectivity as a property of the substance itself and not the skin, thus altering the temperature emitted and recorded by the thermographic imager (Ludwig et al., 2014) and appropriate caution needs to be exercised when setting up experiments and interpreting results. Application of such substances is quite common in pregnancy, as many patients apply cream to prevent the development of stretchmarks. Steketee (1976) looked at various creams and ointments and the effect they had on skin temperature and emissivity. It was shown that there was a difference in temperature when the creams were applied, with each cream and ointment having different temperature changes. More recently, a study showing the effect of sunblock cream on thermographic imaging, demonstrated that the cream application caused a distortion in the normal temperature dissipation over the skin. This is due to the enhanced

contrast between for example skin with superficial vessels underneath, and skin without. The contrast does decrease in time, however the cream application can be an additional factor which alters the thermographic image (Villasen, Sanchez-marin, & Garay-sevilla, 2008). If such cases of topical application of ointment, gel or solution on the skin cannot be precluded then correction of temperature readings are necessary because of the resulting alteration of emissivity. This is necessary because otherwise there is a high risk of obtaining biased measures of temperature, especially where the reflected environmental temperature is very different from that of the objective body (Bernard et al., 2013).

2.2.1.3 In the current research method, the researcher purposely chose not to instruct the patient to refrain from smoking or use of creams prior to the test. The effect of these factors on the relationship between the temperature measured on thermography and the liquor volume on ultrasound could be analysed in the study population. This could subsequently be compared to effects of these same factors described in existing research in non-pregnant patients. Technical Factors

The final group of potentially confounding factors belong to the thermographic equipment itself. There have been a vast number of improvements in thermographic equipment over decades of research, however when it comes to the individual effect thermography may have on research, there are issues that need to be accounted for and may influence the collection of data (Fernández-Cuevas et al., 2015).

2.2.1.3.1 Validity

Validity has been defined as the ‘extent to which a test measures what it is intended to measure’. There are broadly three types of validity; content validity, criterion-related validity and construct validity. With content validity, the researcher is making sure that the multiple factors that make up a questionnaire or scale, are appropriately sampling the general factors

that define the construct that is being measured. With criterion-related validity, the researcher looks at the relationship between the target test and a gold standard test relating to that same construct. Finally, with construct validity, the researcher is making sure that the instrument being assessed is able to measure the theoretical dimensions of a construct, such as observable behaviours (Portney & Gross, 2020). In relation to thermographic research, validity of the test means that the device is able to measure the temperatures of an objects' surface accurately by measuring recorded infrared radiation using a thermal camera (Fernández-Cuevas et al., 2015). Sherman, Wideman, & Karstetter (1996) carried out a study looking at the validity of different forms of thermographic imaging, including video thermography. They showed that video thermography, which is the early precursor of modern day infrared thermography, where the whole image is visualised and not only a spot temperature, had a 98% positive predictive value and a sensitivity of 77%, which was higher than all other modalities. Throughout the literature there are multiple studies which conclusively demonstrate the validity of infrared thermography as a diagnostic tool when used in the context of several pathologies. Chan, Cheung, Lauder, Kumana, & Lauder (2004) demonstrated an increased validity in the use of spot temperature testing of the ear as compared to the forehead, in order to improve temperature detecting systems at airports (Chan et al., 2006). In another study it was shown that a diagnostic accuracy of 74% can be obtained when comparing the frequencies of temperature gradients of cold fingers after a cold test and the Raynaud's diagnosis (Kurt Ammer, 1996). However, there is no specific research that looks at the validity of the use of thermography in the obstetric field. Consequently, This researcher chose to use criterion-related validity of thermography in this study; specifically concurrent validity as indicated in cases where an unproved potential tool is being studied as an alternate, measurement method (Portney, 2020). Concurrent validity is considered when target and standard test measures are acquired more or less concurrently using the potential tool and another previously accepted mechanism so that they both return the same task

outcome. In this study correlations between thermography temperature of a flexion point and the oral thermometer temperature readings where used to establish concurrent validity by carrying out correlations between the temperature readings and the oral temperature taken during the study (at 15 minutes, measurements indicated a significant correlation; $r = .432$, $p > .001$).

2.2.1.3.2 Reliability

Reliability refers to the degree to which a test will give the same result when the measurement is repeated. In quantitative research, reliability tests aim at showing the measure of reproducibility of human observers, laboratory instruments or analytical tests (Lu & Shara, 2007). Reliability reveals both the extent of correlation and also the conformity between measurements (Daly & Bourke, 2008). A measure of reliability should therefore indicate both these criteria (Koo & Li, 2016). Such a consistency index of instruments or analytical methods that is usually utilised to measure reliability is the intra-class correlation coefficient (ICC), which can be a measure of intra-operator and inter-operator reliability (Fernández-Cuevas et al., 2015).

For infrared thermography, reliability of the device is obviously important, and this has been extensively researched, in various several medical specialities. The results of various reliability studies in thermography have been discordant. McCoy et al. (2011) showed excellent inter-examiner and intra-examiner reproducibility of para spinal skin temperatures, whilst the inter-observer reliability was very poor when interpreting mammary thermography in different centres (Mustacchi et al., 1990). Zaproudina, Varmavuo, Airaksinen, & Närhi (2008) carried out reproducibility studies in healthy individuals and found high inter-observer reliability They however found poor intra-observer reliability in the body extremity areas over time (Zaproudina et al., 2008). Silva et al. (2018) showed an ICC value of > 0.75 and

0.70 for intra and inter-reliability respectively for the use of infrared thermography in measuring temperatures of the plantar surface of patients suffering with diabetes.

One study looked at the reliability of thermography in pregnancy and was carried out by Simoes, Vardasca, & Nogueira-Silva (2012), which showed an ICC reliability coefficient ranging between 0.93 and 0.943 at 95% confidence interval. This specifically was looking at the reliability of ROI selection of the abdominal region when analysing the temperatures of the body in late pregnancy.

In this current study, the researcher carried out separate inter- and intra- reliability studies to assess the reliability of thermography when it comes to selection of ROI areas, and thus evaluating the reproducibility and consistency of results obtained during the study.

2.2.2 Camera Factors

2.2.2.1 Distance from Subject

When looking through various protocols of thermographic studies, factors relating to the camera positioning and the distance from the subject of study are mostly clearly defined. The distance recommended however, is determined individually for each specific study that is described, as opposed to converging on a common consensus on the most appropriate single distance that is universally appropriate. A study published in 2006, looked at the operational differences between thermographic cameras which work at wavelengths (λ) of 3-5μm and 8-12μm respectively. It reported that very little temperature correction is required beyond 1m distance from the subject with cameras operating at this wavelength, whilst for cameras operating at 3-5μm wavelength a correction of approximately 0.1˚C for every metre increase is required (Ivanitsky, Khizhnyak, & Khizhnyak, 2006).

The IR-cameras used for the current study where two, the first one was used during the pilot study and operated at 3-5μm λ The camera used in the main experiments (Phases 2

and 3) operated at λ of 7.5 to 13μm. In the light of the above research, the distance from camera to subject throughout the study was kept in the range of 1-1.5m.

2.2.2.2 Height and Orientation

The position of the camera in relation to the subject being studied has also been discussed as being a factor which may influence the thermal images. Mainly, it was found that it is the angle at which the camera is positioned, rather than the height, which may be of relevance (Fernández-Cuevas et al., 2015). In his study, Ammer (2003) examined various positions of image capture, in order to be reproducible and reduce system errors. He found that with temperature measurements of a curved surface, as is the pregnant abdomen, the angle of radiation off the subject and the camera could be a source of false measurements. Small losses in capture of the image are seen starting at a 30° forward azimuth angle from a subject-camera reference, with the losses becoming critical at 60° or more.

For the purposes of the current study, the researcher reduced the angle of the camera in relation to the subject as much as possible, and used the smallest angle that was required in order to obtain the most complete image of the abdomen. This was more so in the Phase 2 study where the protocol was refined subsequent to the findings of the pilot study. However, patient comfort was paramount all throughout the experiments and in some instances the angle at which the patient lay back on the couch had to be altered.

2.2.2.3 Resolution and Sensitivity

One of the differences between IR cameras is dependent on two factors: resolution and sensitivity. Resolution is the camera sensor feature that indicates the detail it is able to acquire and sensitivity defines how accurately the camera can differentiate between intensities of infrared radiation (Etehadtavakol & Ng, 2017). A basic concept of thermal imaging in ‘starting systems’ type cameras is that the image is formed by projection onto all pixels of the detector array simultaneously. The detectors are arranged in a matrix of columns

and rows forming a focal-plane array (Vollmer & Möllmann, 2018). Broadly speaking, each pixel of the thermogram represents one temperature data set, meaning that the more pixels, the higher the resolution and the more thermal information is provided by the image (Fernández-Cuevas et al., 2015). The cameras used for the study had a resolution of 320 x 256 pixels and 320 x 240 pixels for the camera used in the pilot study and Phases 2 and 3 respectively, and a thermal sensitivity of <0.05°C. This pixel resolution is one commonly used in research for medical clinical applications (Fernández-Cuevas et al., 2015), making it an acceptable resolution to use in our study.

2.2.2.4 Image processing and Data analysis

When the thermal image is encoded in the camera memory the thermogram can be principally analysed qualitative and/or quantitatively (Fernández-Cuevas, Lastras, Galindo, & Carmona, 2017). Thermographic cameras without analysis software would just provide qualitative colour patterns as images that can only provide a limited subjective analysis. However, when quantitative outcomes from data analysis of the image are required, software tools are indispensable (Vollmer & Möllmann, 2018). Processing of the image and analysis of the image data are important in medical thermography as it makes thermography more objective and reliable. Temperature data secured from the thermal images can be evaluated and represented by the software as mean numeric values ± standard deviations (Lahiri, Bagavathiappan, Raj, & Philip, 2017).

The software is usually provided by camera manufacturers; however, it is mostly not targeted for use in thermographic studies in humans. A review by R Vardasca, Plassmann, Gabriel, & Ring (2014) looked at over 35 different software packages available, both freeware and licenced, and their individual applications. None of them however were targeted to use in human studies, and suggested that certain systems used by the medical profession

should be integrated into the software so as to be more easily used to analyse thermal images in humans.

The ROI selection is also important when analysing thermal images. Results from an ROI can be assessed against results from contralateral or adjacent ROI's of the same or other subjects. This makes the method of ROI selection controversial if grounded on a subjective manual process because it leads to suboptimal ICC reliability results (Fernández-Cuevas et al., 2017). The Glamorgan protocol was introduced in 2008 aimed at standardizing manual ROI definition in human body thermographic work (Quesada, Kunzler, & Carpes, 2017). It specifically looked at the repeatability of the temperature readings of 90 ROI's, evaluated according to the shape of the ROI. The result was that different shaped ROI's are best used in different parts of the body. For example, an hour-glass shaped ROI was best used at the anterior knee, whilst a circular ROI was best used at the shoulder on the posterior back (Kurt Ammer, 2008). Few studies related to optimal thermographic imaging ROI selection in pregnant women have been conducted. One such study on healthy late pregnant subjects by Simoes, Vardasca, & Nogueira-Silva (2012) already mentioned in section 1.1.6.3, considered ROI's with various shapes, with the right and corresponding left side of the body having similar areas.

The ambiguities of manual ROI selection has led some reviewers to suggest the development of software solutions to provide objective, repeatable selections through an automated process (Fernández-Cuevas et al., 2017).

Indeed, the ROI selection shape is now typically a feature of the analysis software used. The one used in the current study, FLIR®Tools (FLIR Systems Inc., 2020c), provided selectable circular ROI shapes. These where used to best represent four quadrants of the pregnant abdomen, by placing them as close to the median as possible, with least temperature overlap as will be further detailed below.

2.3 Ethical Considerations

Ethics approval was obtained from the University of Malta via the University Research Ethics Committee (Reference Number: FRECMDS_1617_049; Appendix E – Ethics Approval). Each participant was given an information leaflet with information regarding the study and signed a written informed consent form prior to participation. All data pertaining to each participant was kept anonymous and stored in a safe place to be used for research purposes only. The patient had the option to withdraw from the study at any time. The researcher is a Specialist Obstetrician and Gynaecologist and has experience with potential issues that may be encountered with pregnant patients being positioned in such a way for an hour, as well as possible anxieties due to the nature of the study. The researcher could assure all the participants that the technology can be safely used in pregnancy, and could also stop the study if any distress or discomfort was noted. There was no financial remuneration for participation.

The researcher was present at all times whilst the study was being undertaken, and each participant was given the contact number of the researcher to be used both before and after the study should any issues arise or questions need to be asked.

2.4 Study Design

Figure 16 below is a flow chart showing the overall study design. Each phase will be further discussed in the respective segments of the chapter. A reliability study was also carried out and the methodology for this is discussed at the end of the chapter.

2.5 Phase 1 - Pilot Study

2.5.1 Objectives

The literature review highlighted the paucity of studies on the use of thermography in the obstetric field. This lack of research also rendered the creating of an adequate study design to answer the main research questions more challenging. For this reason, it was decided that an initial pilot study was necessary to explore any hypothetical correlation that

might exist between IR thermography temperature recordings and ultrasound depth measurements in healthy pregnant participants. This is the inductive phase of the study and was treated with very broad methodology so as to capture as many findings as possible at this stage and then to refine the next stage design. The aim of this pilot study in Phase 1 was therefore to explore any indicative correlation between the thermographic image data of the pregnant abdomen and the pregnancy characteristics found on ultrasound within the same subject. The direction and strength of the relationship will provide a basis for determining sample size for the next phase. Additionally, any observations with regards to biophysical maternal factors and experiment flow and logistics would provide pragmatic adjustments to the study design.

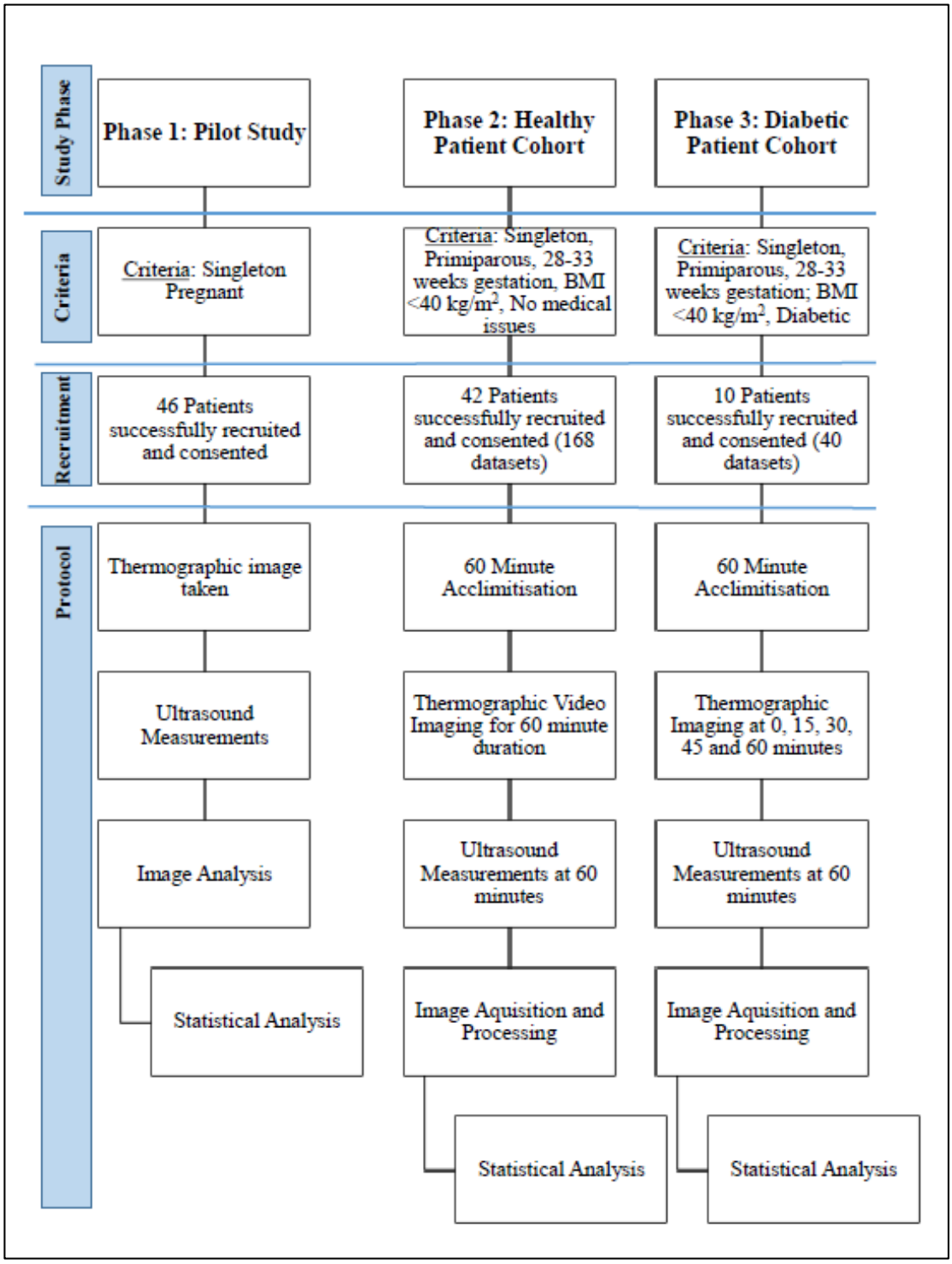


Figure 16. Study design flow chart

The research question for Phase 1 was:

Is there any relationship between the temperature recorded on thermographic imaging and the depth of the liquor pool as measured on ultrasound scan?

2.5.2 Participant Selection and Sample Size Calculation

2.5.2.1 Sample Size

The study design is one based around a correlation study. Correlation is a way to describe the association between two variables. A correlation can be negative or positive depending on the direction of the relationship. A correlation coefficient is what is used to quantitatively describe the strength and direction of the relationship. It is represented by *r* and lies between -1.00 and +1.00; at -1.00 there is a perfect negative correlation, at 0 there is no correlation and at +1.00 a perfect positive correlation exists. Perfect correlations are rare, however the closer the *r* value is to ±1.00 the stronger the correlation is (Portney, 2020). When applying this to hypothesis testing, the null hypothesis for this phase of the study is that the measurements by the two methods are not linearly related and that the correlation is 0 (Bland & Altman, 1986). The alternative hypothesis is usually non directional and is tested using two-tailed tests, when the direction could be positive or negative, or else one-tailed, when the direction is known to be either positive or negative. The measure of the magnitude of the observed effect from the null hypothesis indicates the 'effect size' and this value has an influence on the observable outcome of a test (Field, 2018).

2.5.2.2 Power Analysis and Sample Size Calculation

When planning a study sample, some key factors need to be considered to enable calculation of the sample size. These are the significance level or the acceptable probability of rejecting the null hypothesis (H_0) when in reality it is true (denoted by α), the tolerable probability in not rejecting H_0 when in reality it is false (denoted by β), and the 'effect size' (Daly & Bourke, 2008). α, is conventionally designated at .05, which is arbitrary but generally acceptable (Portney, 2020). Rather than the value of β itself, its complement 1 - β is often used which is the 'power' of the test. Power is the probability that a test will of

attaining statistical significance and lead to rejection of H_0 if actual differences exist (Daly & Bourke, 2008). Typically the minimum target power level used is 0.8 (Field, 2018).

Another factor to consider in estimating the sample size is the prospective effect size value. The effect size for a correlation is the correlation coefficient, *r* itself. Cohen (1992) offered some commonly used proposals for conventional strengths of what represents a large or small effect: $r = 0.2$ (small), 0.5 (medium) and 0.8 (large). For any research study this should be estimated either based on previous research or else on what would be clinically meaningful (Portney, 2020). No previous studies where undertaken correlating the depth of the deepest liquor pool on ultrasound and the temperature of this same pool on thermographic imaging. Consequently the researcher deemed a medium effect size ($r = 0.5$) as reasonable.

An 'a priori' sample size calculation was carried out using the G*Power 3.1.9.4 application (Faul, Erdfelder, Lang, & Buchner, 2007) using the values discussed above and a two-tailed test assumption since the direction of correlation was not yet known. The sample size required for the pilot study was 29 and for a more sensitive power of .95, the size was determined at 46 patients as shown in Figure 17.

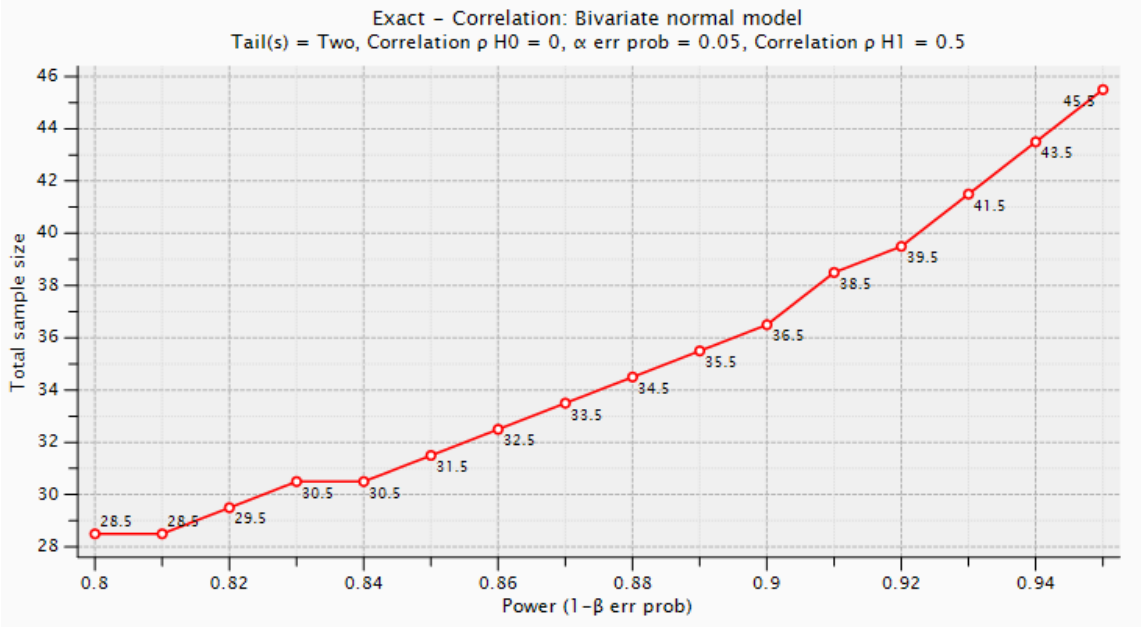


Figure 17. Power - sample size Pilot

2.5.2.3 Recruitment

The participants were recruited as a convenience sample at the Mater Dei General Hospital (Msida, Malta) from out-patients' routine ultrasound sessions. Convenience sampling is a widely used method of nonprobability sampling where subjects are selected on the basis of availability. A pragmatic technique is consecutive sampling which involves recruiting patients who meet the inclusion and exclusion criteria as they become available consecutively to attend a routine screening examination at a clinic (Portney, 2020).

At the time of the pilot study, at the Hospital there were no standard stipulated tests that were carried out during each pregnancy, meaning that the ultrasounds carried out were at the discretion of the obstetrician of care for that particular patient. This also meant that the patients that were on the particular list for the selected days of research were randomly assigned in terms of gestation, maternal conditions and foetal conditions. A set of inclusion and exclusion criteria were therefore established. The patients that fit these criteria, were then given information leaflet regarding the nature of the study and those accepting to participate, were then asked to sign a consent form for the study. A sample of the consent form is seen in Appendix C – Consent.

2.5.2.3.1 Inclusion and Exclusion Criteria

The inclusion criteria where the following:

i. The patient is pregnant. The research is looking at specifically the temperature studies in pregnant patients and thus this is a requirement. The gestation however was not determined during the pilot study in order to maintain a broader approach to the analysis.
ii. The patient is pregnant with a single foetus. Multiple pregnancies could alter temperature measurements during pregnancy, be it from increased

blood flow and larger placenta, to increased foetal effects and increased maternal physiological changes.

In total 46 patients were successfully recruited for the pilot study. This matched the sample size suggested at a power of 95%.

2.5.3 Study Environment

The study was carried out in the designated ultrasound room at the Out Patients' department at Mater Dei Hospital. This room was chosen as it is where the ultrasound equipment is found and these routine ultrasounds are carried out. Outside the room there are chairs where the patients wait for their appointment. The room measures 5.2m x 4.5m and is kept at a steady set temperature of approximately 21°C in view of the ultrasound machine.

The overall layout is shown in Figure 18. Blinds were drawn over room windows in

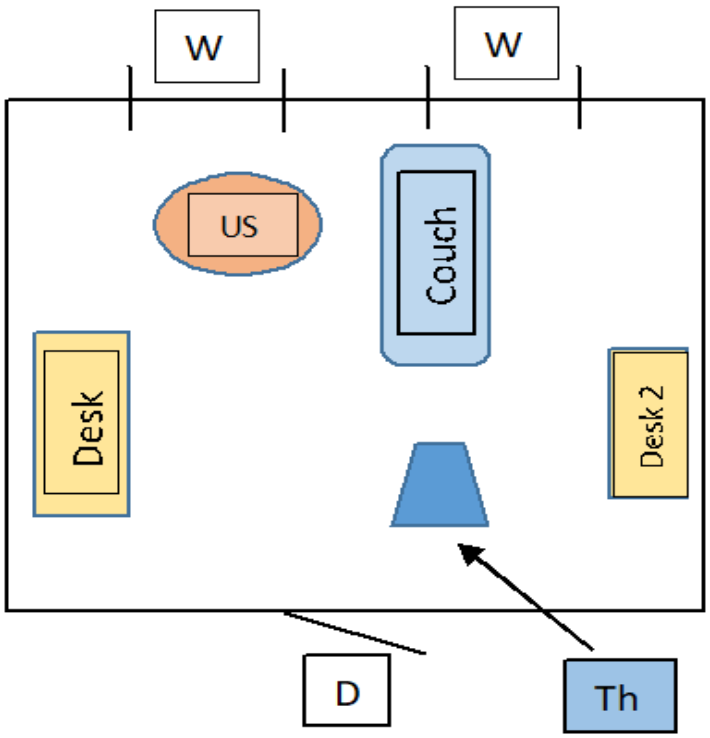


Figure 18. Layout of the Pilot study room

US – Ultrasound; W = Window; D= Door; Th = Thermographic Camera

order to reduce thermal radiation as much as possible. The ultrasonographer was seated in

front of the ultrasound machine (US). The laptop with the thermographic software was placed on the desk and operated by two engineers from the University of Malta. The thermographic camera (Th) was placed on a tripod at the edge of the patient couch at a distance of approximately 1m from the objective. The researcher was sitting down at “desk 2”. The patients’ partner was very often present and was seated on the right hand side of the couch.

2.5.4 Equipment

Image acquisition was done using a FLIR SC7200 thermal camera shown in Figure 19.

This camera, has a spatial resolution of 320×256 pixels a temperature resolution of 0.02°C and works in the 3-5µm wavelength infra-red range (FLIR Systems Inc., 2020b). The camera was placed on a tripod during the experiment and was connected to a power supply so as not to rely solely on the battery life and prevent unexpected loss of data. The camera was also connected to a laptop running the FLIRtools (FLIR Systems Inc., 2020c) software which allowed images to be recorded remotely. An emissivity of 0.98 and a reflectivity of 0.02 were

Figure 19. FLIR SC7200 Thermal Camera

set in the camera. The focal length of the thermographic camera was such that the acquired image included the entire abdominal region. These images where saved on the same laptop hard drive for subsequent analyses. An example of the image that would be captured is shown in Figure 20.

The ultrasound machine used to carry out the ultrasound images was an Esaote myLab ™ X5, which is the machine utilised routinely for the out-patients ultrasound appointments at the Hospital. A curvilinear probe was used to carry out the ultrasound and measurement relating to the pregnancy.

2.5.5 Procedure

The patient was consented before the routine ultrasound appointment. The

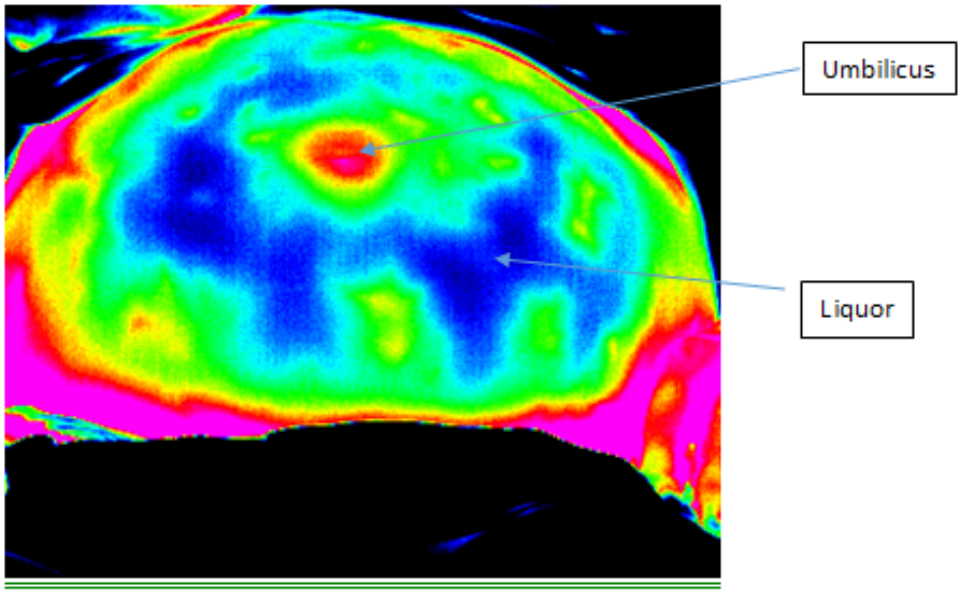


Figure 20. Typical thermographic image capture

thermographic camera and laptop were set up prior to the ultrasound session started and images were only recorded once the consented patient was in the ultrasound room and positioned on the couch.

Not all patients having an ultrasound were participants in the study, however the thermographic set up was not dismantled between patients and non-participating patients

were assured that no images were being recorded. In these cases all personnel related to the research waited outside the room, whilst the ultrasound examination was being carried out. When recruited patients were called in for their appointment, they were joined by the researcher and the engineering personnel who helped operate the equipment.

The patient was asked to lie supine on the examination couch. The couch is set at an incline angle of 45 degrees. It is important that the patient is not placed in a totally flat position on the couch as this can lead to Caval Compression Syndrome. This is a condition which occurs beyond 20 weeks' gestation, where in the supine position the uterus compresses the inferior vena cava. This reduces the blood flow from the extremities to the maternal central cardiac system and leads to symptoms of low blood pressure. These can vary from maternal dizziness and fainting, to foetal bradycardia (low heart rate).

Once the patient is on the couch, they are asked to expose their abdomen. The abdomen needs to be exposed from under the breast region, to just above the anterior superior iliac spines as shown in Figure 21. The room door is closed and locked in order to assure as much privacy as possible.

The thermographic image was then captured remotely using the FLIRtools (FLIR Systems Inc., 2020c) software. After the thermographic image is taken, the ultrasonographer proceeded to carry out the routine ultrasound. It was requested by the research team, that the initial readings to be taken by ultrasound before carrying out a full assessment would be the following:

i. The depth of the liquor pools in four quadrants, starting from the right lower quadrant, followed by the right upper quadrant, followed by the left upper quadrant and finally the left lower quadrant.

Figure 21. Exposed abdomen

ii. The position of the foetus, meaning if the foetus was in cephalic (head down) position, breech (feet down) position or transverse position.

iii. The placental location – whether this was anterior of the uterus, posterior of the uterus, at the upper fundal part of the uterus, or low down in the uterus.

iv. The thickness of the abdominal wall at and around the umbilicus. Five readings were taken at ultrasound, these being the depth at the umbilicus, at 3 o'clock, at 6 o'clock, at 9 o'clock and at 12 o'clock

These measurements being taken in this order, before the routine scan is carried out, does not pose any risk to the patient or the foetus. A typical ultrasound image of the four quadrants is shown in Figure 22.

2.5.6 Data Analysis

2.5.6.1 Thermographic Data Analysis

The FLIRtools (FLIR Systems Inc., 2020c) software was used to extract data from the thermographic image with all the individual images being processed in the same way. From

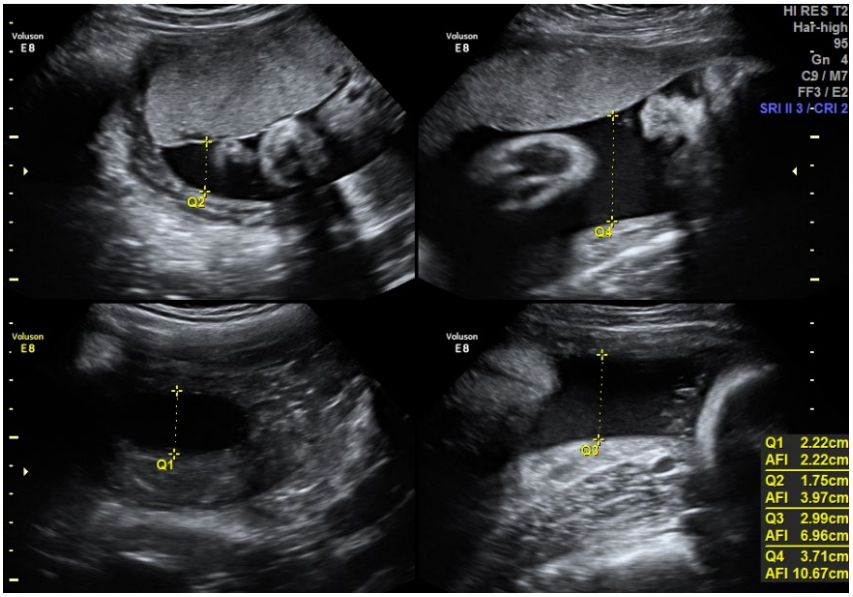


Figure 22. Typical ultrasound image showing the four quadrants
(Henry Knipe, Radiopaedia.org)

the thermographic data a graphical representation using a rainbow palette, with red as hot and blue-black as cold was obtained and assessed qualitatively. The thermographic software can allow 'alteration' of the temperature range resulting in an 'alteration' in the visible 'colour', making the coldest and hottest spots more visible to the operator. The following measurements were recorded:

i. The site of the coldest area on thermographic image

ii. The temperature of the coldest spot

iii. The temperature at umbilicus

2.5.6.2 Statistical Analysis

The measurement data obtained from the thermographic and ultrasound images was compiled under the respective variable headings (Appendix A – Data fields). The data was assessed for normality using a QQ Plot and was shown to be normally distributed as shown in Figure 23.

A bivariate Pearson product-moment correlation analysis was conducted between depth of the deepest liquor pool and the temperature of the coldest thermographic area across

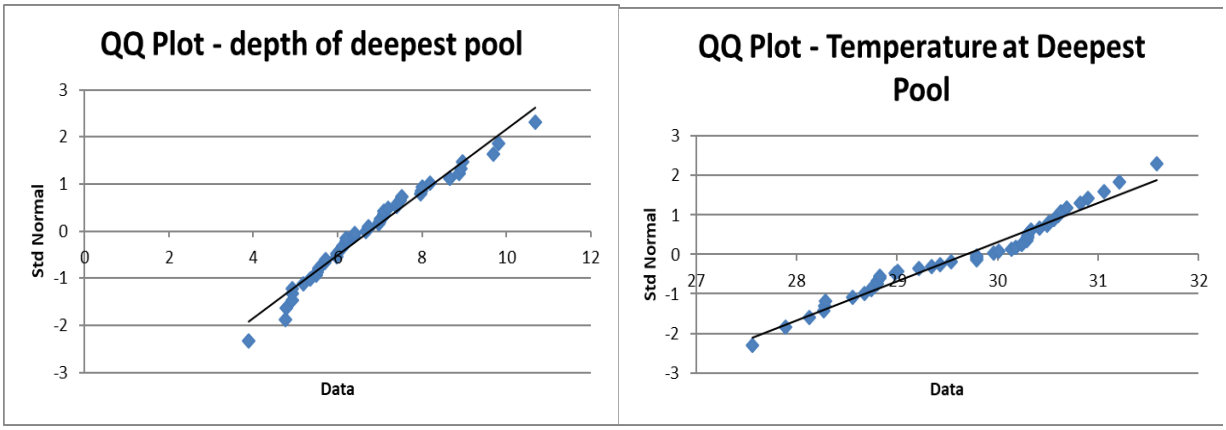


Figure 23. Q-Q plots pilot data

participants using SPSS (IBM Corp, 2010). The null hypothesis (H_0) and alternative hypothesis (H_1) of a two-tailed significance test for correlation was represented as:

H_0: $\rho = 0$ (there is no significant correlation between the two variables)

H_1: $\rho \neq 0$ (there is significant correlation between the two variables)

2.5.6.2.1 Outcome

A statistically non-significant correlation between the two variables was obtained; r = -.24, p = .099, 95% CI [-.543, .077]. This value of r gives an indication of the potential effect size and will be used to determine sample sizing for Phase 1.

The objective of this phase was to garner information to be able to enhance the design of the study proper. A variety of qualitative factors were recognised which were incorporated in the study process. These are described in the phase 2 Methodology, section 2.6.1 below.

2.6 Phase 2 – Healthy Cohort

2.6.1 Overview

The protocol for Phase 2 was refined with information and insights gained from the pilot-study so as to improve the quality of the data that was collected. A choice to look at healthy pregnant woman was taken so as to create a baseline of readings relating to thermography and to ultrasound findings, whilst taking into account biophysical maternal

factors and assessing how these can alter the relationship between the two modalities. Also, more emphasis was made on researching the time required for thermographic imaging temperature to reach a steady state in the pregnant patient. All affected changes in procedure are summarised in Table 2.

Factor	**Pilot-study**	**Phase 2**	**Rationale**
State of health	Few exclusions	Only healthy participants	To create a data baseline
Acclimatisation	Not specified	Fixed extended period	To determine optimal point for data acquisition
Region of Interest (ROI)	ROI was whole abdomen	Abdomen evaluated as 4 ROI quadrants	This matched the data delivered by ultrasound measures and provided more data points
Power Analysis	An arbitrary effect size of 0.5 and two-tailed test	An effect size of 0.24 as found from pilot study and one-tailed test from correlation results of pilot	This allowed for a more accurate Power calculation
Gestational age	No exclusions	Between 28 and 33 weeks	Avoided difficulties with thermographic measurements due to curvature of abdomen in late gestation
BMI (Body Mass Index)	No exclusions	<40 kg/m^2 at start of pregnancy	Minimise problems with excess weight affecting thermographic imaging
Parous status	No exclusions	Primiparous	Eliminate possible temperature variation due to uterine muscle mass, blood flow changes or scaring in multiparous patients at the abdominal site
Study environment	Used room at the Out Patients' department	Used room close to the obstetric wards	New room less prone to thermal interference (such as windows), more private and patient friendly
Thermographic camera	FLIR SC7200	FLIR E60	The FLIR E60 is lighter and more portable
Operators	Researcher + ultrasonographer + thermal camera operator	Researcher only	Improve thermal interference profile and enhance patient privacy

Table 2. Changes to procedures following pilot phase

2.6.2 Objectives and Research Question

The aim of Phase 2 of the study was to assess the use of thermography in a group of healthy pregnant women with specific inclusion and exclusion criteria which will be described in the following section.

The research questions to be answered by this phase of the research were:

i. Is there a significant relationship between the thermographic temperature level and the maximum depth of the liquor pool measured with ultrasound at the regions of interest (ROI's) in a healthy pregnant patient? If there is, can this be quantified?

ii. Does an acclimatisation period prior to thermographic measurements improve relevance of readings? What is this optimum acclimatisation period?

iii. Do maternal and biological parameters influence any correlations and how do these intrinsic and extrinsic factors affect this relationship?

2.6.3 Research Design

A prospective quantitative study was carried out, so as to analyse the correlation between the measured depth of liquor pool as seen on ultrasound and the measured temperature of the respective liquor pool as measured by thermographic camera. The data was collected over a period of 60 minutes, at 15 minute intervals in a controlled ambient temperature, so that the acclimatisation pattern can be analysed for this group of patients. Biophysical properties and demographics pertaining to the patients were collected and applied to the analysis of the data, as well as properties pertaining to the utero-foetal unit.

2.6.4 Participant Selection and Sample Size Calculation

2.6.4.1 Sample Size

The result of the pilot study showed a Pearson correlation coefficient of -0.24 between the depth of the deepest liquor pool observed on ultrasound and the temperature of the same pool observed on thermography. The second phase study was to focus on the relationship between the liquor pool depth observed on ultrasound and the temperature of that same pool on thermographic imaging. For this reason, the researcher used an r = -0.24 to calculate the sample size required for this second phase using G*Power (Faul et al., 2007) software. Using a one tailed a priori analysis, with a level of significance α of 0.05, power of 0.9 and a null hypothesis of zero, the sample size was calculated to be 145 as shown in Figure 24.

For each patient studied, four quadrants of data were collected. Since each of this data was an objective and independent measurement it meant that in order to obtain a 145 case sample, the researcher would have to recruit a minimum of 37 patients (37 x 4 = 148 data sets).

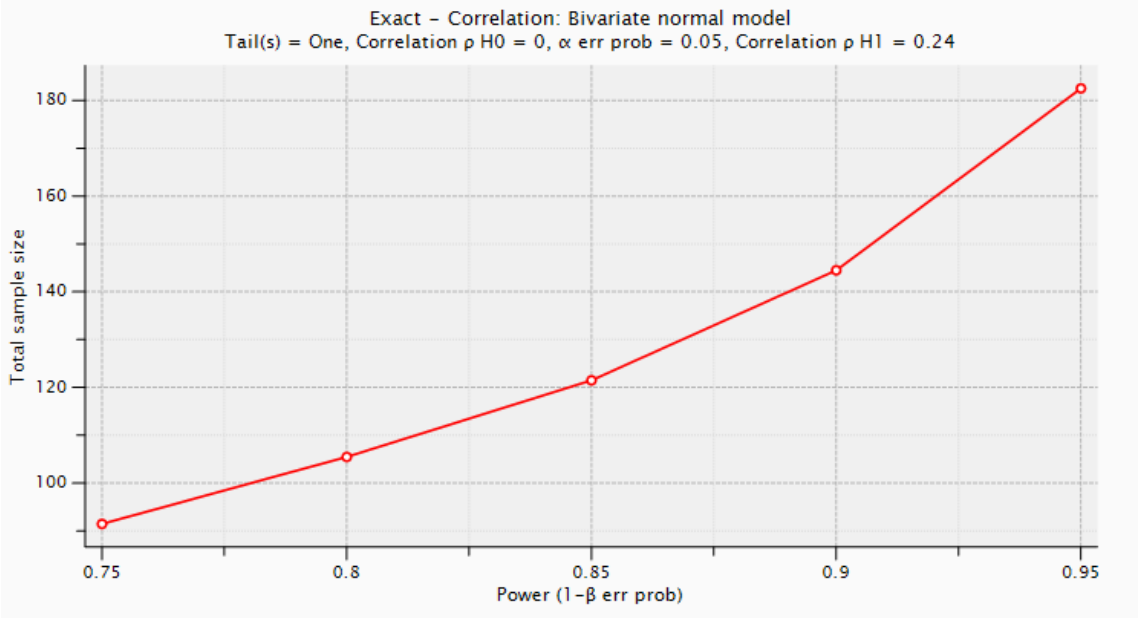


Figure 24. Power - sample, Phase 2

2.6.4.2 Recruitment

The recruitment source of patients was from the appointment list for routine O&G departmental 'Anomaly scans'. These ultrasound scans are carried out on every patient who books her pregnancy at Mater Dei Hospital – if she so wishes. It is usually carried out between 21 and 23 weeks' gestation. The list typically comprises most of the pregnant women in Malta at any one time. It therefore provides for a representative pool for sample selection.

When booking the routine appointment each patient was handed an information leaflet about the study and if interested the patient was asked to complete a study-interest form. The information requested on this form included the patients' name, email and telephone number. The patient was informed that she would be contacted by the researcher in due course.

The interested patients' list was screened against the acceptance criteria and also to specifically exclude from the study those patients in the following categories:

i. Patients who were listed as having an ultrasound for fertility reasons. This would include those listed as 'IVF', 'Detailed' or 'Gynae' in the records.

ii. Patients who were shown to have had previous admissions to labour ward, an obstetric ward or breast-feeding clinic in the preceding years, suggesting that this was not their first pregnancy. Any patient who was seen to have any of these admissions was assumed a multiparous patient and thus was excluded from the study.

iii. Patients who were noted to have 'twin pregnancy' as an indication for the ultrasound were excluded from the study.

The remaining patients were presumed to be nulliparous, thus being eligible for the study. These were then contacted individually over the telephone as per contact details previously provided.

On calling the patients, the researcher introduced herself as a Resident Specialist in the Department of Obstetrics and Gynaecology of Mater Dei Hospital and thanked them for their interest in partaking in the study and for agreeing to be contacted. The study objectives were briefly explained and further information regarding participation was offered if required. All the patients were interested to hear more about the study and requested more information. This was subsequently sent to them via email (Appendix B – Patient Communication) and they were encouraged to respond to the email declaring their interest or lack of as soon as possible. It was also confirmed that the patient was in fact primiparous and carrying a singleton pregnancy. The estimated due date for the delivery was also requested. Once the woman agreed to participate, a date was scheduled to carry out the study.

Quantitatively, this process yielded the following participant numbers. An independent recruiter recruited 286 patients over the course of 12 months. Each of these patients was given an information leaflet explaining the research and the procedure. They were then contacted by the researcher at a later date. 172 patients contacted refused to participate in the study after reading the information leaflet and having all questions answered. 57 patients did not fit the selection criteria mainly because they were not primiparous or did not fit the gestational age criteria. 14 patients, although agreed to participate, could not attend on particular study days in view of other commitments. Overall, 43 patients participated in the study. The flow chart in Figure 25 depicts the sample recruitment process.

In this way a convenience sample comprising a total of 43 patients was successfully recruited. Unfortunately, one patient had a fainting episode during the study and so was not included, leaving 42 patients for data analysis.

Figure 25. Sample recruitment process

Fig xx – Flow chart demonstrating the data collection for Phase 2 study

2.6.4.3 Inclusion and Exclusion Criteria

The following are the criteria the patient was required to have in order to participate in the study.

i. The patient had to be pregnant

ii. The pregnancy had to be her first pregnancy. This will eliminate any possible temperature alterations that may occur due to uterine muscle mass

or blood flow changes present in uteri which have previously carried a pregnancy. Also, any scars caused by a previous caesarean section could also affect the temperature changes at the abdominal site.

iii. The gestation was of between 28 and 33 weeks' gestation. This is an arbitrary gestational age. During the pilot study, where there was no gestational age restriction, it was seen that there was a difficulty in capturing the thermographic image of the upper abdominal quadrants in cases of late gestation. This is due to the seated position of the patient and the resulting curvature of the abdomen. With the thermographic camera on the tripod, it is difficult to capture these regions of interest. It was therefore decided, that earlier gestations have smaller curvatures of the abdomen, meaning that the full abdominal curvature can be adequately captured with the thermographic camera on the tripod and the patient at 45-degree angle in the sitting position.

iv. The pregnancy had to be a singleton pregnancy. Multiple pregnancies could alter temperature measurements during pregnancy, be it from increased blood flow and larger placenta, to heightened foetal effects and compounded maternal physiological changes.

v. The patient has a pregnancy which is classified as a low risk pregnancy.

Patients were excluded from the study if they satisfied the following criteria.

i. The patient was suffering from obstetric complications such as gestational diabetes, intra-uterine growth restriction or placental conditions. These complications could alter the temperature regulation during the pregnancy by altering the blood flow within the placenta and umbilical cord, resulting in alterations of temperature regulation and thus beyond the scope of the research.

ii. The patient had a BMI (Body Mass Index) of ≥40 kg/m2 at the start of the pregnancy. This extreme of weight was chosen as a cut-off for exclusion in the experiment since it has been shown to affect thermal patterns in body areas(Fernández-Cuevas et al., 2015).

2.6.5 Study Environment

The setting used for the pilot study had various issues that where improved upon during the Phase 2 of the research. A room close to the obstetric wards within Mater Dei Hospital was chosen as the study setting. Its location is ideal as it is near to a large waiting area; also it is familiar to patients and easier to find. The selected room was the ultrasound room used for ward patients and free to use every day after clinical hours. Also, as shown in Figure 26, it was smaller than the room that was used in the pilot study, at 4.61m by 3.49m. It

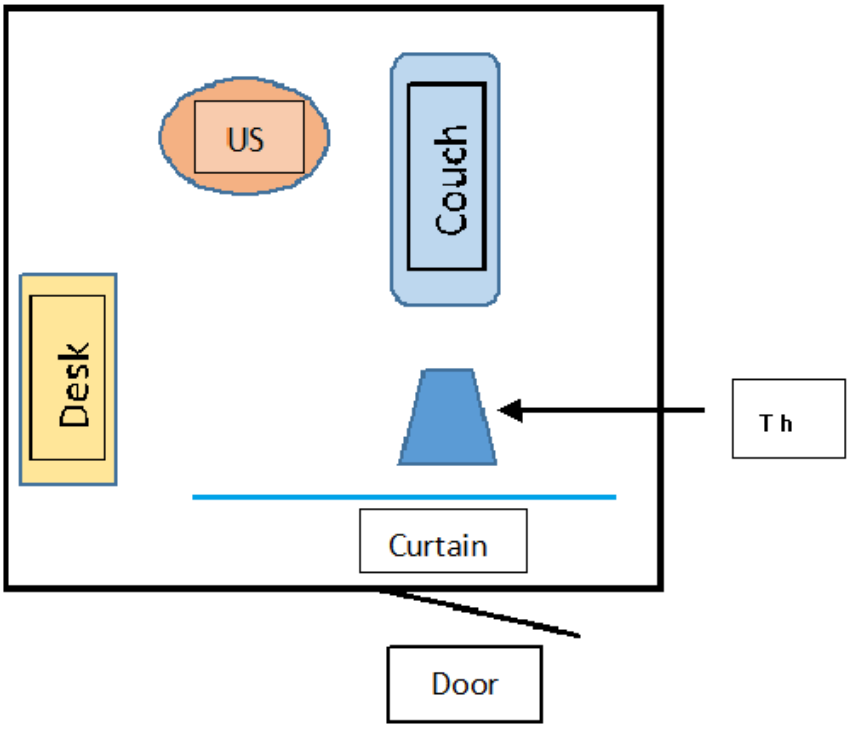


Figure 26. Study setting used for Phase 2

US = Ultrasound machine; Th = Thermographic equipment

had no windows and had a privacy curtain shielding the patient during the experiment.

The temperature and humidity level where set by the Hospital's central environment control system and the main room lighting was standard ceiling fluorescent lights. The desk lamp and the computer where switched off during the experiment to reduce any thermal radiation interference. The ultrasound machine was also switched off during the thermographic image acquisition to reduce any ambient temperature alterations.

Another improvement in Phase 2 over the pilot study was to reduce the operating personnel involved with the experiment to have a thermally 'cleaner' ambience. Only the patient and researcher would be in the room with the researcher conducting the ultrasound examinations as well as the thermographic image acquisition process.

2.6.6 Equipment

The thermographic camera used for Phase 2 was a FLIR E60 (FLIR Systems Inc., 2020a) which is a lighter and more portable camera. The camera is shown in Figure 27. This camera has an infrared resolution of 320 x 240 pixels, with a thermal sensitivity of < 0.05˚C and a temperature accuracy of 2˚C. The spectral range is 7.5 to 13µm (FLIR Systems Inc., 2020a). The camera was connected to a laptop directly with the FLIRtools software used to record the images as video as well as single images.

The ultrasound machine used during the experiment was an Esaote Mylab™ X5 (Esaote S.p.A, 2020). This machine was routinely utilised by the ultrasonographers during day clinics.

2.6.7 Procedure

The patient was requested to sign the consent form (Appendix C – Consent) which they would have already read through the information leaflet given to them on recruitment, and any questions and concerns answered. The consent form was filed securely.

Figure 27. FLIR E60 Thermal Camera

The Phase 2 study involves an acclimatisation profile, which was carried out over 60 minutes. For this reason, the patient is offered the opportunity of emptying her bladder prior to the start of the experiment. This is both for comfort reasons, but also to avoid bladder distension which could compress abdominal contents including the uterus, as well as creating another source for heat production. The patient is then asked to lie on the couch in a supine position at a 45-degree angle. As discussed previously, this is carried out so as to avoid caval compression syndrome. The patient is made as comfortable as necessary, including placing multiple pillows behind her head, or even lower back. The researcher is constantly mindful of the fact that the patient must be in this position for just over 60 minutes with minimal movement. The patient is then asked to expose her abdomen from just under the breast area, to just below the hips.

The FLIR E60 camera is mounted on the tripod, and connected to the laptop where the FLIRtools software is running. The camera is also connected to the electrical input and a

full battery is ensured in the camera. The tripod is placed at the edge of the couch at a distance of approximately 1.5m from the patients' abdomen. The angle of the camera is adjusted according to the individual patient so that the whole abdominal curvature is visualised on the live feed. The setup is shown in Figure 28.

It is important to note that the annual calibration maintenance of the thermographic cameras used during this experiment were managed by the Biomedical Cybernetics Department at the University of Malta. Since the cameras were used by different users, it was important to make sure that before each participant, the emissivity was set at 0.98 and the distance at 1 metre, to standardise the temperature measurements taken for each participant.

Prior to the start of the experiment, a photograph of the patients' abdomen is taken in order to document any scars, moles, birthmarks, striations, as well as documentation for skin type.

The FLIR camera is set to record a video at 7.5 fps (frames per second) via the FLIR software on the laptop and once the patient is comfortable, the recording is started. The patient is asked to minimise movement from side to side, avoid passing her hand in front of her abdomen and to avoid touching her abdomen.

Whilst the thermal imaging video was being recorded, the patient was asked questions regarding her pregnancy as per a pre-set questionnaire (Appendix D – Questionnaire). Data was collected pertaining to the pregnancy itself, including the gestation and any medication being taken. Data was also collected regarding the patients' smoking habits over the past 3 days, if any oils or creams were used on the day of the experiment and any previous surgeries carried out in the abdomen prior to, or during, the pregnancy. The patients' blood pressure and pulse were taken using a digital blood pressure monitor. This was done on an outstretched arm so as not to create movement and interfere with the thermographic recording of the abdomen. The patients' oral temperature was then taken using a digital thermometer. Again, this does not interfere with the thermographic image of the abdomen being recorded.

At 55 minutes, the ultrasound machine was switched on and the patients' details are entered into the system, as is routinely done for all the ultrasounds taken in the hospital setting. At 60 minutes, the thermographic video is stopped and a still thermal image was acquired.

Figure 28. Setup for Phase 2 study

The patient was asked not to move until the ultrasound is carried out. The researcher carried out the ultrasound after placing ultrasound gel on the patients' abdomen. A number of measurements were taken in the following order - foetal lie, right lower quadrant liquor pool depth, right upper quadrant liquor pool depth, left upper quadrant liquor pool depth, left lower quadrant liquor pool depth, placental location and abdominal circumference. A linear probe was then used to measure the abdominal thickness at 3, 6, 9 and 12 o'clock positions relative to the umbilicus.

All ultrasound images were transferred to and stored in a portable drive for later analysis. Finally, an individual patient report is printed at the end of the ultrasound. Since the foetal movement could displace the liquor, it is important to minimise the time between the 60 minute still frame on thermography and the ultrasound measurements of the liquor pools.

The patient was helped to sit up slowly being mindful of the fact that the patient might feel dizzy and so this should be done at her own pace. Finally, the patient has her height and weight measured so as to calculate her BMI.

2.6.8 Data Analysis

2.6.8.1 Thermographic Data Analysis

Thermographic data analysis was carried out using the FLIRtools (FLIR Systems Inc., 2020c) software. In order to reduce any bias the thermographic images were analysed after the ultrasound is carried out.

The thermographic video file data was extracted for each participant and file frames at 0, 15, 30, 45 and 60 minutes were analysed. The emissivity parameter was set to 0.98 prior to the start of the analysis. The following markers and Regions of Interest (ROI's) were located on the frame image as below:

i. An ROI labelled EI1 is placed over Right Lower quadrant

ii. An ROI labelled EI2 is placed over Right Upper quadrant

iii. An ROI labelled EI3 is placed over Left Upper quadrant

iv. An ROI labelled EI4 is placed over Left Lower quadrant

v. A point temperature marker labelled Sp1 is placed over the umbilicus

vi. A point temperature marker labelled Sp2 is placed over a fixed point on the wall behind the patient

vii. A point temperature marker labelled Sp3 is placed over a flexion point where this was visible

For each ROI, the software identifies the maximum, minimum and average temperature. The four thermographic ROI's are placed to most closely represent the four liquor quadrants which were measured on ultrasound at 60 minutes. Since the thermographic image is a video stream, the patient might move slightly between one reading and another, and for this reason the ROI's and Sp1, Sp2 and Sp3 might require manual adjustment to better locate and synchronise the wanted regions accordingly. A typical analytic representation is shown in Figure 29.

All fields of data that were recorded from the thermographic analysis, ultrasound measurements and questionnaire are listed in Appendix A – Data fields.

2.6.8.2 Statistical Analysis

A database of all participants was compiled showing the thermographic data for each quadrant and for the umbilicus at the selected five time-points measured in °C and ultrasound measurements at the 60-minute point with pool depth being measured in cm. Additionally, computed fields factored the temperature difference between the wall temperature and the quadrant at the set time-points and at 0 minutes. The demographic and bio-medical parameters of each patient were included.

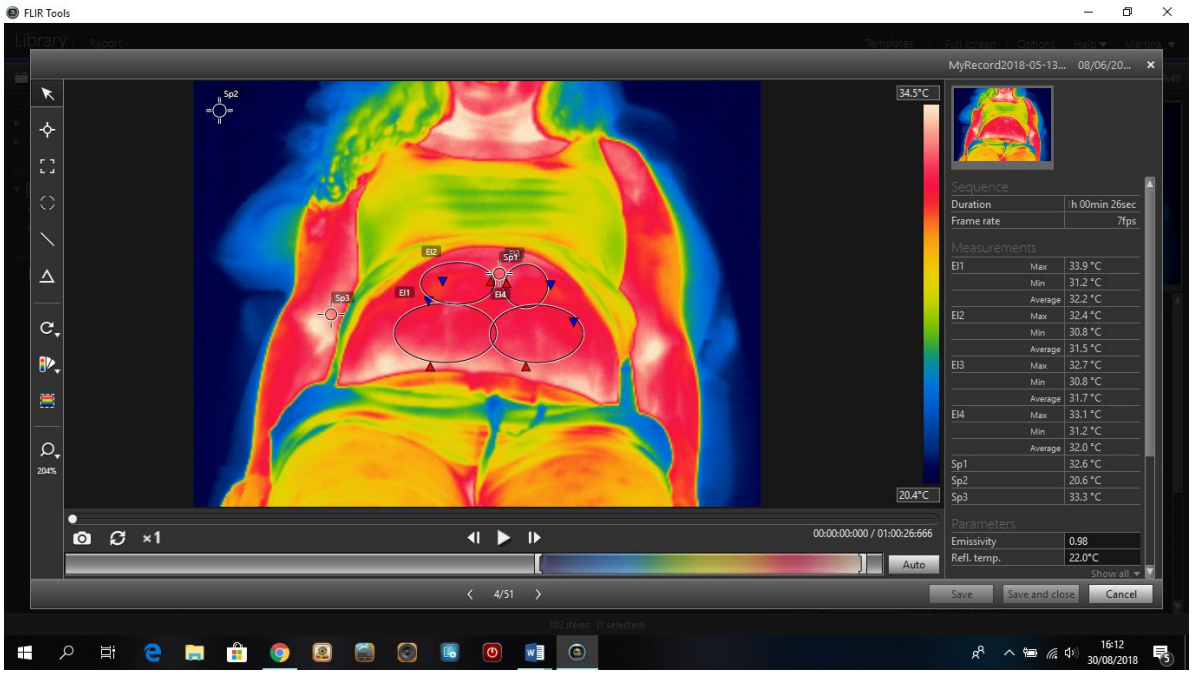


Figure 29. Typical Phase 2 thermographic data analysis

Data is screened for outliers, correctness and normality. Being normally distributed, parametric tests could be used for analysis. A mixed ANOVA analysis is used to explore any main effects on thermographic data over time and location. By comparing time-point group means significant acclimatisation time-lines can be indicated. Also comparing quadrant location group means helps determine dependence of temperature readings on location.

Bivariate Pearson product-moment correlation analysis is conducted to assess any statistically significant relationships between liquor pool depth measured on ultrasound and the temperature readings measured thermographically for each of the measured and computed variables.

For statistically significant correlations of temperature factor and pool depth, a moderation analysis is conducted to assess any interaction between the predictor (temperature measure) and moderation variables (bio-medical factors) that affects the outcome (pool depth measure).

Finally a regression analysis is done to determine the best linear model based on the available data that can serve to describe and qualify the relationship between variables that help predict outcome values and pool depth. This is done by carrying out a backward stepwise multiple regression. The main aim of a regression model is to predict the outcome or explain the relationship between the variables that are being analysed and this method, where variables are removed or retained depending on how they maximise the prediction accuracy with the least number of predictors, results in the smallest number of independent variables that would make the most valuable contribution to predicting the dependant variable (Portney & Gross, 2020).

2.7 Phase 3 – Diabetic Cohort

2.7.1 Overview

During this phase of the study, the researcher aimed to repeat the study protocol carried out in Phase 2, this time within a clinical group, as opposed to a healthy pregnant group of patients. This group of choice was pregnant patients suffering from diabetes. The study made no distinction between type 1, type 2 and gestational diabetics.

There have been numerous studies showing the way in which IRT can be applied to this area of medicine successfully. However, the highlight of this past research is mainly focused on the more advanced forms of the disease displaying signs of neuropathy and vasculopathy (Lahiri et al., 2012), These advanced stages are very rarely seen in the obstetric population, mainly because these are a younger cohort of patients.

Research involving the study of thermoregulation of early stage diabetics has shown that the physiology of a diabetic patient is different to a non-sufferer. In cases of type 1 diabetics, the few studies related to whole body exposure to heat, showed that the core temperature largely remained unaltered. It has been shown however that patients in this subgroup of the condition tend to be in a general hyperinsulinemia state which results in a moderate vasodilation in the skin and extremities, which results in an increase in heat loss

compared to a non-diabetic individual. It was also shown that there are changes within the morphology of the blood vessels which affects the way they can carry out thermoregulatory mechanisms. With type 2 diabetics, studies have also shown a similar effect in the modulation of vasodilation and impaired skin blood flow responses secondary to temperature changes. Glycaemic control across all types of diabetes is altered in hot ambient conditions by increasing the insulin absorption within the body. Some studies have also shown that there are differences of up to 3% in glycaemic levels between the winter and the summer months due to the ambient temperature (Kenny, Sigal, & Mcginn, 2016).

2.7.2 Objectives and Research Question

This study intended to analyse if the findings that resulted in the Phase 2 study with healthy patients could be replicated in a group of patients suffering with diabetes in pregnancy and if not what differences arise. In view of the physiological changes that occur within this group, this could shed some light on how and if thermography can be a useful analytical device for a pregnant diabetic cohort.

The research questions asked in this part of the study are:

i. Is there a significant relationship between the thermographic temperature level and the maximum depth of the liquor pool measured with ultrasound at the regions of interest (ROI's) in a patient suffering from diabetes during pregnancy?
ii. Do maternal and biological parameters influence any correlations and how do these intrinsic and extrinsic factors affect this relationship?
iii. Is there a difference between the temperature and pool depth relationship in the healthy patient group and in the diabetic patient group?

2.7.3 Research Design

This Phase 3 study had similar objectives and design as that of Phase 2. The results from this sub-group of patients were then compared to the larger group of patients analysed in Phase 2 of the study.

2.7.4 Participant Selection and Sample Size Calculation

2.7.4.1 Sample Size

The results of Phase 2 of the research showed a significant correlation between the maximum quadrant temperature less the wall temperature at 30 minutes and pool depth. The Pearson's correlation coefficient *r* was found to be 0.529 and $R^2 = 0.28$. In this next stage, the researcher aimed at investigating these findings in a clinical group, specifically pregnant patients suffering from diabetes. In order to calculate the sample size, this R^2 value was used in an a priori power analysis, using G*Power (Faul et al., 2007) set for linear multiple regression F test. With a level of significance of 0.05, power of 90% and one predictor, the sample size was calculated as 30. The sample size to power graph is shown in Figure 30.

2.7.4.2 Recruitment

The patients were recruited from the diabetic pregnancy clinic held fortnightly at Mater Dei Hospital. This is a clinic where diabetic pregnant patients are referred from general obstetric clinics in order to have specialised obstetric care by a specialist team which includes an obstetrician with an interest in diabetes and endocrine conditions, a diabetologist and a dedicated midwife. These patients would either be patients with pre-existing type 1 diabetics or type 2 diabetes or would have been diagnosed with gestational diabetes during their routine obstetric visit by having fulfilled set criteria as described by the unit guidelines.

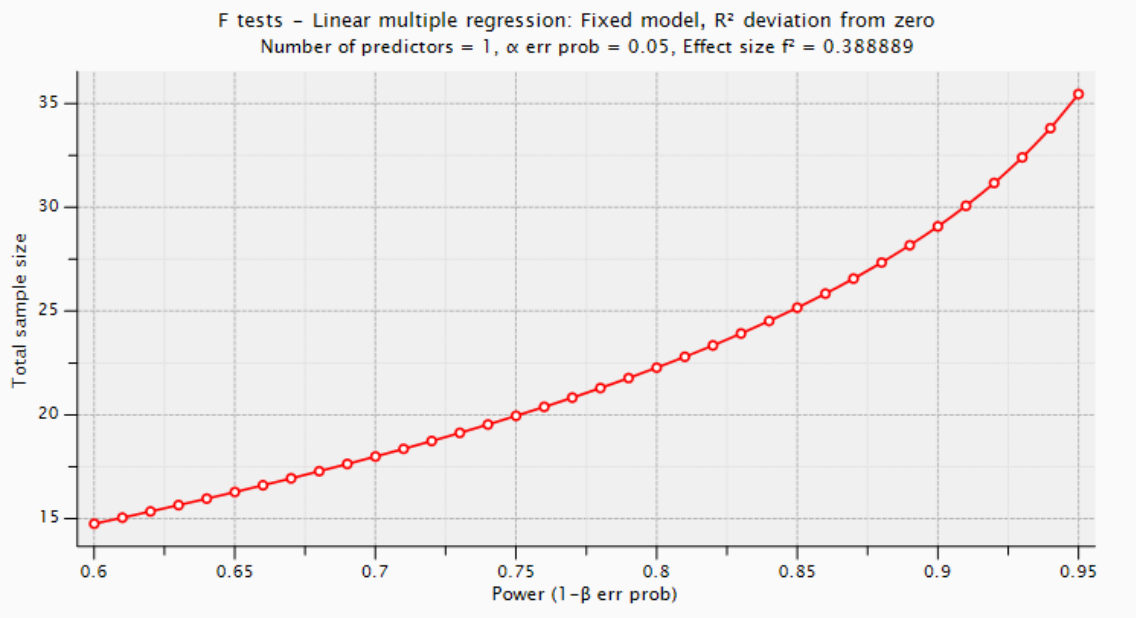


Figure 30. Power - sample, Phase 3

The recruiters where fellow obstetrician colleagues who run the diabetic clinic. They were given a set of inclusion and exclusion criteria and if the patient fulfilled these criteria, they were asked if they would be interested to participate in the study. If they were in fact interested they were given an information leaflet and subsequently contacted by the researcher and the processes continued as for Phase 2.

According to the National Obstetric Information System (NOIS) report of 2017, 4.4% of all the deliveries that year (4325) were diagnosed with diabetes – be it pre-existing type 1 or type 2 diabetes, or newly diagnosed gestational diabetes. The number of primiparous mothers recorded during that year was 2274 – 52% of all deliveries (Gatt & Borg, 2018). The relatively small proportion of diagnosed diabetic pregnant patients yearly, the alternate weekly set up of the clinics, and the expected level of patients not willing to be recruited or fail the selection criteria where all factors that suggested potential difficulties to recruite the required population numbers within the timeframe of the study.

If one assumes a 4.4% diagnosis of pre-existing or gestational DM in the current antenatal screening system, only about 90 potentially recruitable patients are available

annually for recruitment. With a participation acceptance rate of 10%, recruitment time of a sufficient sample could take over a year to acquire.

Ultimately, 10 patients were recruited which nominally provided 40 data cases, which fulfil the power requirements, calculated above.

2.7.4.3 Inclusion and Exclusion Criteria

These were essentially similar to those of Phase 2 with the exception that here inclusion was conditional on the patient suffering from diabetes in pregnancy.

2.7.5 Study Environment

The study setting was the same as that used in the Phase 2 study, which was preferable so as to maintain stable conditions across the studies.

2.7.6 Equipment

The thermographic camera and ultrasound machine that was used in Phase 3 was the same as was used in Phase 2 of the study.

2.7.7 Procedure

The researcher followed a process mostly similar to that of the Phase 2 study. In Phase 3 however, the researcher chose to take still thermographic images of the patient at 0, 15, 30, 45 and 60 minutes rather than a thermographic video. This was done in view of the results observed in Phase 2, where analysis of the data recorded at 15 minute intervals proved to be sufficient. The patient was notified prior to each image being taken and asked to minimise movement from side to side, avoid passing her hand in front of her abdomen and to avoid touching her abdomen during this time.

The patient was asked the same questions regarding her pregnancy as in Phase 2 and the data was collected in the same way. The same ultrasound measurements were carried out and stored for analysis.

2.7.8 Data Analysis

2.7.8.1 Thermographic Data Analysis

The individual images were processed in the same way as the video frames in the Phase 2 study using the FLIRtools (FLIR Systems Inc., 2020c) software set at the same emissivity of 0.98 prior to each analysis. Similar ROI's are selected as in Phase 2 and were adequately documented and stored. All fields of data that were recorded from the thermographic analysis, ultrasound measurements and questionnaire are listed in Appendix A – Data fields.

2.7.8.2 Statistical Analysis

A database of all participants was compiled showing the thermographic data for each quadrant and for the umbilicus at the selected five time-points measured in °C and ultrasound measurements at the 60 minute point with pool depth being measured in cm. Computed fields similar to Phase 2 were added and the demographic and bio-medical parameters of each patient included. A combined data set of Phase 2 and Phase 3 participants was also created with a new dichotomous variable indicating clinical condition. This is used to carry out comparative statistical analyses of data from the two cohorts.

Data screening for outliers and correctness is carried out, however as the Phase 3 sample size is relatively small, special attention is given to evaluating that the assumptions of normality, linearity and homoscedasticity are fulfilled. Additionally, where possible in SPSS, 'bootstrapping' is used in the analyses as a mitigating method for reducing any normality bias.

Similar to Phase 2, a mixed ANOVA analysis is used to explore any main effects on thermographic data over time and location. ANOVA analysis of the combined database compares means for the two conditions.

Bivariate Pearson correlation analysis is used to assess any statistically significant relationships between liquor pool depth measured on ultrasound and the temperature readings measured thermographically for the measured and computed variables.

Using the combined dataset of diabetic and non-diabetic participants, a moderation analysis is conducted to statistically assess if any interaction between the predictor (temperature measure) and diabetes condition has any influence on the relationship between thermographic temperature records and pool depth.

A simple linear regression analysis is carried out to suggest the linear model based on the available data that can best depict the relationship between thermographic readings and pool depth for the diabetic cohort.

2.8 Reliability Assessment

For every assessment tool that is researched, it is important to assess its reliability. The reliability of a device or test is defined as the extent to which the measurements can be replicated. An adequate measure of reliability should show both the degree of correlation and the degree of agreement between the measurements. The index that can achieve this is the Intra-class correlation coefficient (ICC). It was first introduced by Fisher as a modification of the Pearson correlation coefficient. Modern ICC is calculated by mean squares which are obtained through the analysis of variance (Koo & Li, 2016).

The results of various reliability studies in thermography have been discordant. McCoy et al. (2011) showed excellent inter-examiner and intra-examiner reproducibility of para spinal skin temperatures, whilst the inter-observer reliability was very poor when interpreting mammary thermography in different centres (Mustacchi et al., 1990). Other research carried out on reproducibility studies in healthy individuals found high inter-observer reliability, however found poor intra-observer reliability in the body extremity areas over time (Zaproudina et al., 2008).

In this study, the researcher wanted to evaluate the intra-rater and test-retest reliability of thermographic imaging of the pregnant abdomen. The intra-rater reliability shows the variation between two or more raters measuring the same sample group. The test-retest reliability shows the variation of the measurements taken by the same observer under the same conditions at different points in time.

2.8.1 Intra-rater Test

Two obstetricians working at the Obstetrics and Gynaecology Department at Mater Dei Hospital, where selected as independent raters (Operator 1 and Operator 2). They both had no prior experience with thermography. They were given a series of random 'practice' thermographic images of the pregnant abdomen, and were asked to select ROI's on the image, using the FLIR software. The ROI's were selected from the patients' right lower quadrant (EI1), following through to the right upper quadrant (EI2), then left upper quadrant (EI3) and left lower quadrant (EI4). These ROI's corresponded to the regions defined in Phases 2 and 3 and as shown in Figure 31. The raters were asked to set the emissivity to 0.98 on each thermographic image.

Once a rater felt confident with the use of the software, they were asked to carry out the same exercise on 15 thermographic images randomly pre-selected by the researcher. There was no time limit on the exercise. Both raters carried out the exercise under the same conditions, on the same laptop and in the same room.

The data was extracted from each thermographic image for the respective rater and compiled manually by the researcher. The measurement for minimum temperature within the ROI, maximum temperature within the ROI and average temperature within the ROI where tabulated as separate variables. SPSS was used to carry out a reliability analysis using a two way mixed model (Model 3,1) Interclass Correlation Coefficients (ICC), absolute agreement at 95% confidence interval. The ICC interpretation score was that as described by Fleiss

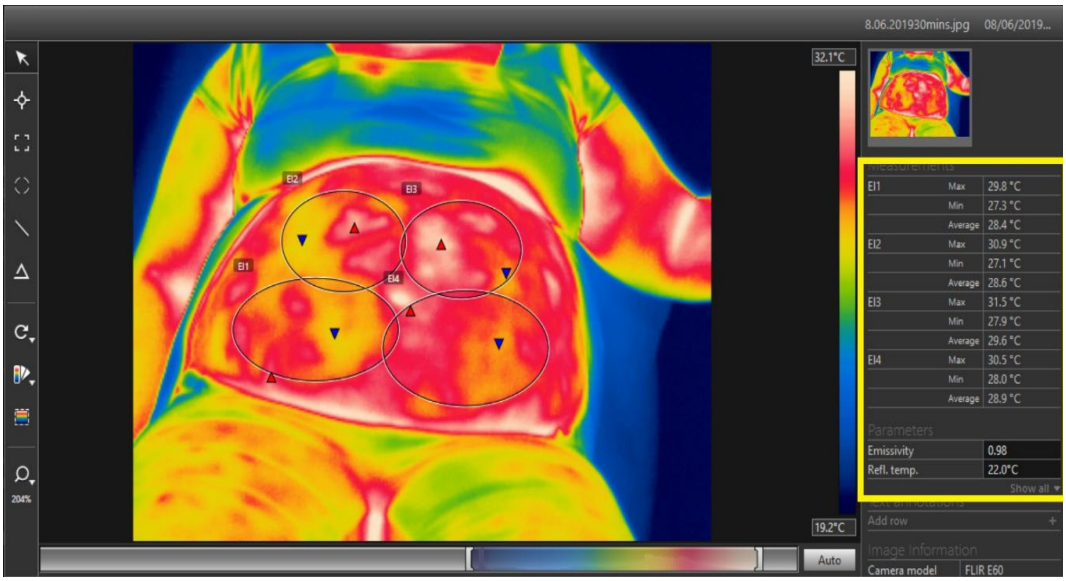


Figure 31. Typical ROI's selected by raters

(2011), namely that 0.4 indicates poor, 0.4 - 0.75 indicates fair to good and 0.75 indicates excellent agreement.

The Cronbach's alpha is a measure of internal consistency of a test. The internal consistency of a test is a measure of the reliability of the test based on correlations of the results. Ideally, the index of Cronbach's alpha should fall between 0.7 and 0.9 – lower correlations may signify that different traits are being measured, whereas higher correlations may signify that the test is redundant (Portney & Gross, 2020). The Cronbach's alpha was carried out using SPSS for the minimum, maximum and average temperature readings within the ROI's comparing the readings of both operators respectively.

2.8.2 Test Re-Test

The researcher selected the ROI's labelled EI1, EI2, EI3 and EI4 as was required of the other two observers, with the same corresponding anatomical landmarks described previously. This was carried out on 15 randomly pre-selected thermogr*a*phic images. This was then repeated 4 days later under the same conditions, using the same thermographic images.

The data was manually extracted from each thermographic image for the respective test and the data for minimum, maximum and average temperature within the ROI was tabulated separately for each test. A reliability analysis using SPSS was carried out as a two way mixed model (Model 3,1) Interclass Correlation Coefficients (ICC), absolute agreement at 95% confidence interval. The ICC interpretation score was as per Fleiss (2011).

The standard error of the mean (SEM) was calculated using the following formula,

$$SEM = SD\ x\ \sqrt{1 - ICC}$$

in order to assess the absolute reliability index of the test-retest reliability. The SD was taken as the average of the SD of the two operators at the respective reading – minimum, maximum and average. The values measured by the researcher are assumed to be the precise values; however, there is potential error in the measurement. The calculation of the SEM provides a more accurate evaluation of the degree of confidence within which the true value lies. The confidence in the measurement lies with a high ICC and the SEM to reflect a limited range (Portney & Gross, 2020).

The minimal detectable change (MDC) was also calculated for the two tests carried out by the same researcher using the following formula,

$$MDC = 1.96\ x\ SEM\ x\ \sqrt{2}$$

The MDC is the minimal measured amount before one can eliminate the possibility that the measurement error is alone responsible for the change. At 95% confidence interval, it means that 95% of readings will show a random variation of less than this amount when the test is carried out on multiple occasions (Portney & Gross, 2020). The above formula takes into account the 95% confidence interval by the multiplication of 1.96 within the equation. A procedure which does not show good test-retest reliability will have a large MDC, meaning that a larger change in readings is required in order to exclude a measurement error as a sole

source of the error itself (Portney & Gross, 2020). The outcome of these reliability tests is reported in the Results section.

3 Results

In this chapter, the results obtained from the research carried out by using the methodology discussed in the previous section, will be described. The results of Phase 2 and Phase 3 of the research will be presented and this will include both the relative descriptive and inferential statistics. The outcome of the Pilot study was included in the previous chapter as its intent was to enhance the methodology for subsequent phases. As described previously, the aim of the initial study was to test a broad methodology design in order to then refine this further to be used in the larger studies. The correlations found as a result of the pilot study, served as a guide and proof of concept to moving forward with the next phases.

The research carried out in Phase 2 and Phase 3 of the study aimed at answering the following research questions:

i. Is there a relationship between the thermographic temperature level and the maximum depth of the liquor pool measured with ultrasound at the regions of interest (ROI's) in a pregnant patient? If there is can this be quantified?

ii. Does an acclimatisation period prior to thermographic measurements improve relevance of readings? What is this optimum acclimatisation period?

iii. Do maternal and biological parameters influence any correlations and how do intrinsic and extrinsic factors affect this relationship?

iv. Does the same relationship between the thermographic temperature level and the maximum depth of the liquor pool measured with ultrasound at the ROI's exist in patients with a known clinical condition in pregnancy, namely diabetes?

3.1 Phase 2 - Overview

The specific aim of Phase 2 of the study was to explore the relationship between the thermographic temperature level and the maximum depth of the liquor pool measured with ultrasound at the regions of interest (ROI's). Additionally, it aimed to investigate if maternal, biological and environmental parameters influence these correlations if they exist.

The first segment of this chapter presents the descriptive statistics relevant to the demographic, biological, ultrasound and thermographic measurements.

3.1.1 Demographic and Biological Measurements

3.1.1.1 Maternal age and Gestational age

These are depicted in Table 3. The maternal age of the participants ranged between 20 and 41 years whilst the gestational age ranged between 27+5 and 31+1 weeks. The mean maternal age was rounded to the nearest whole number, and the mean gestational age was rounded to the nearest gestational day.

	Maternal Age (years)	**Gestational age (weeks)**
Maximum	41	31+1
Minimum	20	27+5
Mean	**29.75**	**28+3**
SD	**5.11**	**1**

Table 3. Maternal age and Gestational Age

3.1.1.2 BMI

Table 4 shows the Body mass index for each patient participating in the study was calculated by measuring the height (cm) and the weight (kg). The mean, maximum and minimum BMI are tabulated below, with the values rounded to the first decimal place.

	BMI (Kg/m2)
Maximum	39.2
Minimum	18.9
Mean	**28.7**
SD	**4.95**

Table 4. Body Mass Index

3.1.1.3 Skin Type

For each patient, the skin type was recorded and quantified according to the Fitzpatrick skin typing scale (Fitzpatrick, 1988) – see Table 5. In the cohort, there were 3 patients with a type 2 skin type, 30 patients with a type 3 skin type, and 9 patients with a type 4 skin type. There were no patients with type 1,5 and 6 skin types in the recruited group.

Skin Type	**Typical Features**	**Patients in Cohort, *N* (%)**
I	Pale white skin, blue/green eyes, blond/red hair	0 (0%)
II	Fair skin, blue eyes	3 (6.97%)
III	Darker white skin	30 (71.43%)
IV	Light brown skin	9 (20.93%)
V	Brown skin	0 (0%)
VI	Dark brown or black skin	0 (0%)

Table 5. Skin type

3.1.2 Abdominal Features

For each of the recruited patients, the presence or absence of striations was documented, as well as whether they used cream over their abdomen within 12 hours of the procedure.

3.1.2.1 Striations

Out of the 42 participants, 5 had visible striations on their abdomen.

3.1.2.2 Use of Cream

Out of the 47 participants, 7 admitted to using cream on their abdomen within 12 hours of the procedure. In all cases, the cream used was for the purpose of stretch mark prevention.

3.1.3 Biological Parameters

3.1.3.1 Blood Pressure and Pulse

For each patient, the Blood Pressure and Pulse was documented using an electronic calibrated Blood pressure machine – Table 6. None of the patients required a large cuff in order to record the data. The mean values where rounded to the nearest integer.

3.1.3.2 Temperature

The patients' oral temperature reading was taken via the same electronic thermometer with sterile cover over the mouth piece. The mean values where rounded to the first decimal place. The ambient temperature the individual day of each procedure was documented for each patient. This was recorded from the Malta International Airport local weather forecast (Malta International Airport, https://www.maltairport.com/weather/). The mean temperature value was rounded to the first decimal place. Table 7 shows data.

	Systolic Blood Pressure (mmHg)	**Diastolic Blood Pressure (mmHg)**	**Pulse (bpm)**
Minimum	161	87	103
Maximum	100	50	57
Mean	**120**	**69**	**85**
SD	**12.28**	**7.96**	**10.26**

Table 6. Blood pressure, pulse

	Oral Temperature (‘C)	**Day Temperature (‘C)**
Minimum	37.3	34.0
Maximum	35.7	14.0
Mean	**36.6**	**24.3**
SD	**0.26**	**5.30**

Table 7. Temperatures

3.1.4 Ultrasound data

The ultrasound data collected from each patient is presented. The following data was specifically analysed for each cohort:

i. Liquor pool depth (per quadrant) (cm)

ii. Placental location – anterior, posterior or fundal

iii. Foetal lie – cephalic, breech or transverse

iv. Mean abdominal thickness (cm)

3.1.4.1 Liquor pool Depth

The liquor pool per quadrant was measured, meaning that each patient had 4 readings which related to the same respective four quadrants on thermogram (RLQ, RUQ, LUQ and LLQ).

As seen in the scatter plot (Figure 32), there is a wide range of pool depth measurements on ultrasound. The numbers range between 9.73 to 0. Since no patient recruited suffered from anhydramnios, the zero values are indicative of void results for that particular quadrant.

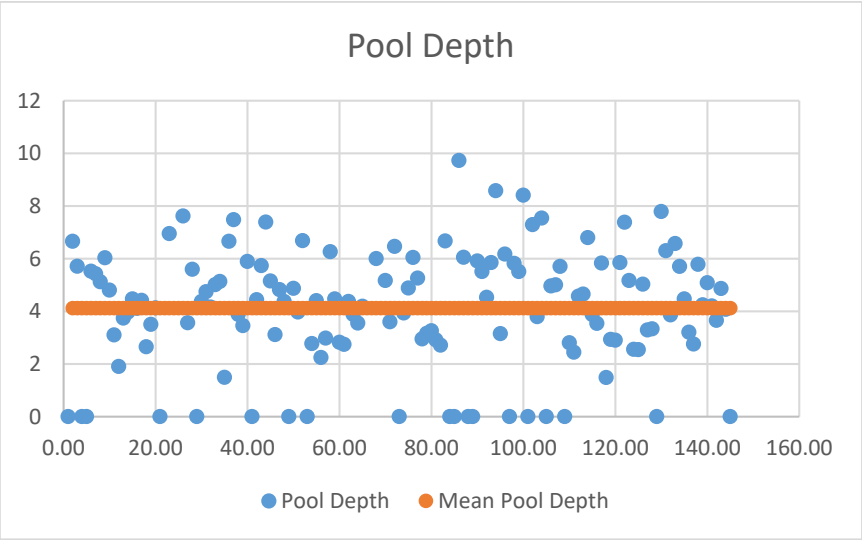


Figure 32. Pool depth scatter plot

Figure 33 shows plotted the DVP of the individual patients and as one can see, no patients have a DVP of more than 10 cm (the upper orange line) which represents polyhydramnios or the upper limit of liquor volume. The lower orange line represents the lower limit of liquor volume at 2 cm and none of the readings fall below this, meaning that no patients suffered from oligohydramnios.

3.1.4.2 Placental Location

The placental location on ultrasound was recorded for each patient. 8 patients had a fundal placenta, 17 patients had a posterior placenta and 17 patients had an anterior placenta.

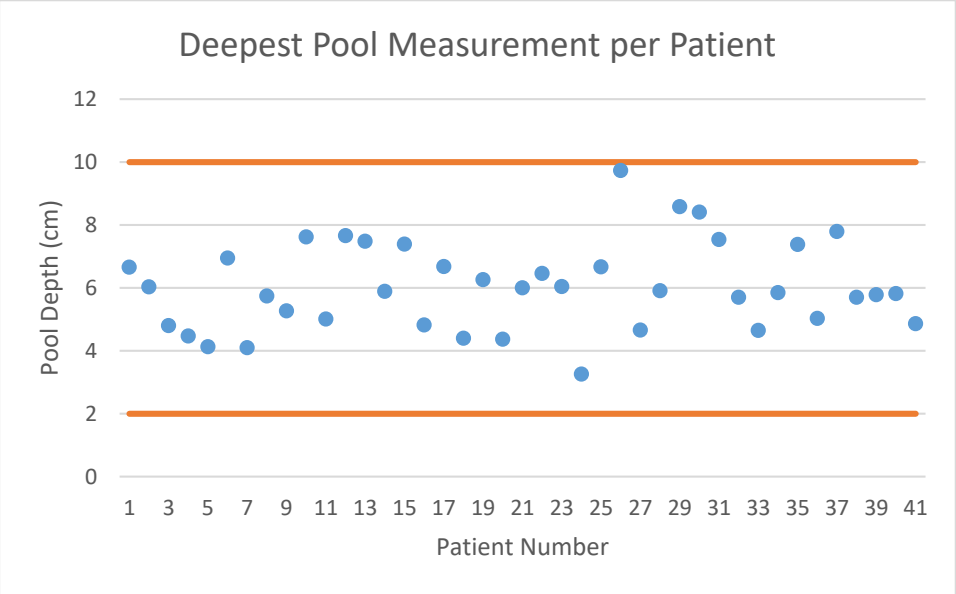


Figure 33. Deepest Pool scatter plot

3.1.4.3 Foetal Lie

The foetal lie on ultrasound was documented on ultrasound, with 36 being cephalic, 3 being breech and 2 being transverse. The foetal lie was not recorded for one of the 42 patients.

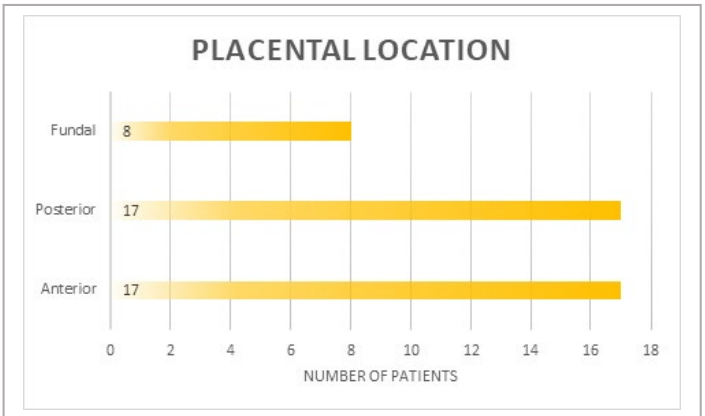


Figure 34. Placental location frequencies

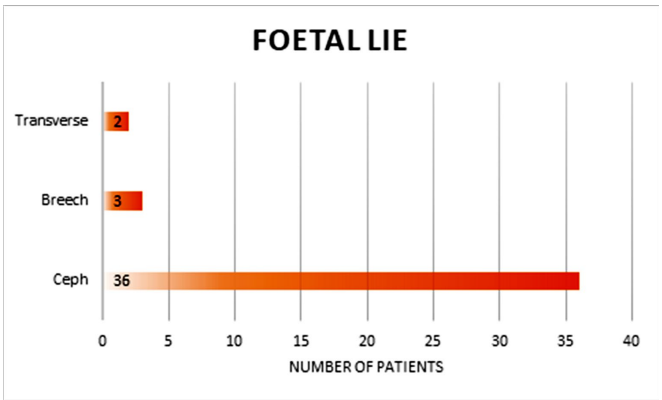


Figure 35. Foetal lie frequencies

3.1.4.4 Mean abdominal thickness

The abdominal thickness was measured by ultrasound by measuring the thickness at the umbilicus 3, 6, 9 and then at 12 o'clock. The mean values of these readings were then calculated for each patient with results shown in Figure 36 (data for missing patient numbers was inaccurate and therefore excluded).

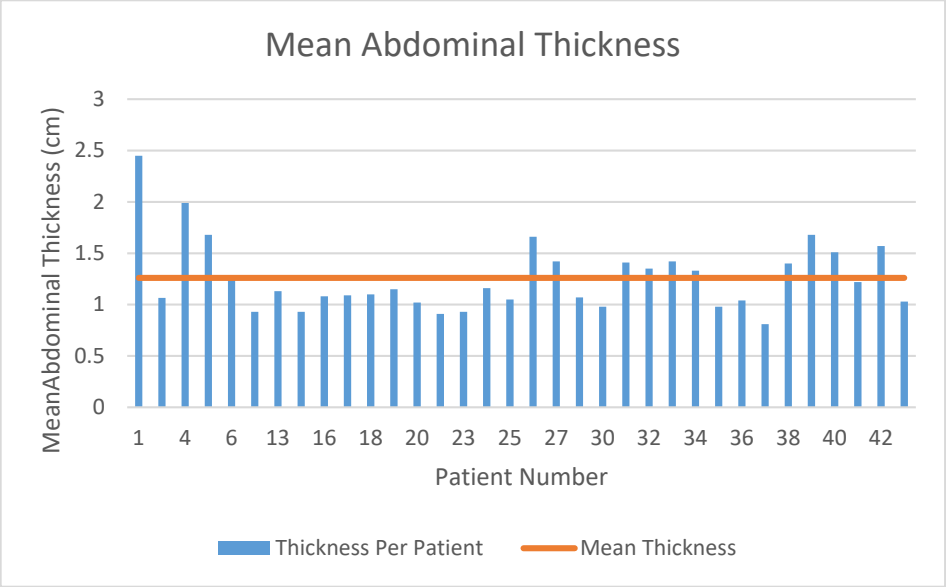


Figure 36. Mean abdominal thickness per patient (SD=0.34)

The next part of the chapter describes the rationale and results of the statistical analyses undertaken. Firstly, the discussion regarding the statistical analysis for screening the data is presented. Following this, the specific tests and their results will be discussed.

3.2 Phase 2 - Data screening

The collected data compiled per subject was screened for accuracy and consistency to identify and resolve possible problems prior to running the data analyses. This is recommended practice to ensure that the reported results are not biased by inconsistencies of accuracy, missing data and outliers in the participants' data (Warner, 2013). Subsequently, the data was assessed to determine how it complied with the assumptions common to general linear model statistical analysis, namely: additivity, linearity, homogeneity or homoscedasticity of variance and independence (Field, 2018). Accuracy and missing data was analysed by examining the range, mean and number of cases for each measure from the descriptive statistics of the dataset (Pallant, 2013). Any values that appeared discrepant were considered for correction if the raw data so justified or resulted in deletion (Tabachnick & Fidell, 2013).

When specifically analysing the data collected for pool depth, since the study sample excluded patients with potential complications, any data outside the wider range 2 to 10 cm

(as per typical guidelines (e.g., NHS University Hospitals Plymouth, 2018)) was considered incorrect and the data excluded. Univariate outliers in continuous variables of interest were detected by computing Z-scores and identifying cases with standardised values greater than 3 standard deviations (for $p < .001$) (Field, 2018). This yielded 5 outlier data points across all the variables of interest which specific points were excluded. Additionally, multivariate outliers analysis revealed two cases with a Mahalanobis distance greater than the χ^2 value for $p < .001$ (Tabachnick & Fidell, 2013). These cases were omitted.

The resultant screened datasets for the participant group whose trial pool depth data was utilized in the analysis is N = 114, $M_{\text{pool depth}} = 4.79$ cm, $SD_{\text{pool depth}} = 1.47$, $Range_{\text{pool depth}} = 2.24 – 8.58$ cm.

Conservative significance tests ($p < .001$) for levels of skewness and kurtosis were used to test for univariate normality (Field, 2018). Only skewness of systolic blood pressure and mean abdominal thickness were found to be statistically significant. However, looking at the shape of their distributions and because of the large sample size (N > 100) it was indicative that these variables do not vary sufficiently from normality to considerably change the statistical analyses (Tabachnick & Fidell, 2013). When univariate normality is sustained for individual variables, multivariate normality can typically also be assumed since in these cases, deviations from normality are usually negligible (Hahs-Vaughn, 2016). This was also confirmed by examining the multivariate P-P plot of the residuals for a random distribution.

Linearity and homoscedasticity were assessed by inspection of a scatterplot of the standardised values of these residuals *zresid* against the standardised values of the predicted outcome *zpred*. There was no indicated systematic relationship between these and the results were reasonably evenly distributed in around the origin supporting the assumptions of linearity and homoscedasticity of the data (Field, 2018).

3.3 Phase 2 - Statistical results

To explore the general question for this study, namely whether a relationship exists between the thermographic image data and the ultrasound pool depth information, an initial analysis of the thermographic data collected to find any significant effects of time and ROI was carried out. Following this, a correlation analysis between the thermographic data and pool depth was undertaken. The results are reported in the following sections.

3.3.1 Significance of group differences

To explore any main effects on thermographic data over time and quadrant location, a 3(temperature - minimum, average, maximum) x 5(time – after 0, 15, 30, 45, 60 minutes) x 4(quadrant location – right upper quadrant RUQ, left lower quadrant LUQ, left lower quadrant LLQ, right lower quadrant RLQ) mixed ANOVA was conducted. Using Greenhouse-Geisser correction for sphericity there were significant main effects of temperature level and time point and their interaction.

For the main effect of temperature level the Greenhouse-Geisser estimate for the departure from sphericity was ε = 0.59. This main effect was significant $F(1.17, 117.1) = 1444.79$, $p < 0.001$, $\eta^2 = 0.94$ which means that if all other factors are ignored, the mean temperature recorded was significantly different between temperature levels. Contrasts revealed that mean temperatures for minimum level, $F(1, 100) = 1584.51$, $p < 0.001$ and average level, $F(1, 100) = 1218.43$, $p < 0.001$ were significantly lower ($M_{min} = 29.94$, $SD_{min} = .82$), ($M_{avg} = 30.95$, $SD_{avg} = .86$) than that for maximum level ($M_{max} = 32.37$, $SD_{max} = .93$).

The main effect for time point was also significant on temperature with the Greenhouse-Geisser estimate for the departure from sphericity ε = 0.40, $F(1.59, 158.9) = 65.75$, $p < 0.001$, $\eta^2 = 0.4$ which means that disregarding other factors, the mean temperature recorded was significantly different between time points. Within subject contrasts showed that mean temperatures for 0 minutes - $F(1, 100) = 80.69$, $p < 0.001$, 15 minutes - $F(1, 100) = 22.05$, $p < 0.001$ and 30 minutes - $F(1, 100) = 12.28$, $p = 0.001$ were significantly higher than that for 60 minute time point. However, temperatures for the 45 and 60 minute time points

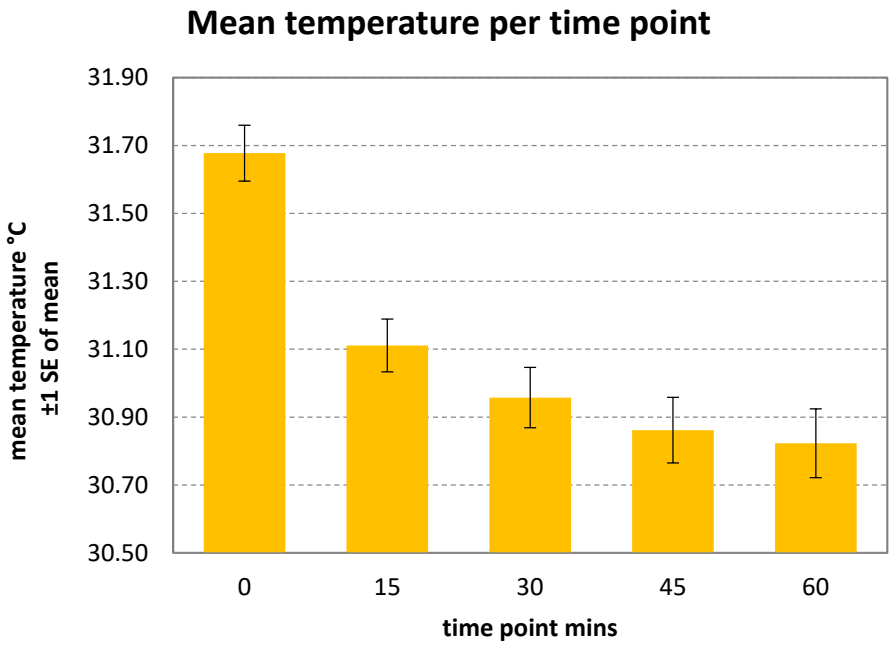


Figure 37. Mean temperature per time point

were about the same across the temperature levels - $F(1, 100) = 1.8$, $p = .183$. As can be seen in Figure 37, the estimated marginal means indicate the cooling or acclimatisation being more pronounced in the initial period falling from $M_{t=0} = 31.68$ to $M_{t=15} = 31.11$ and less marked at later time points $M_{t=45} = 30.86$ to $M_{t=60} = 30.82$.

There was no significant difference between-subjects effect of quadrant location, indicating that recoded temperatures were similar between quadrants, $F(3, 100) = .104$ $p = 0.957$, $\eta^2 = 0.003$. The relevant marginal means are plotted in Figure 37.

The interaction of temperature level with time point was significant with the Greenhouse-Geisser estimate for the departure from sphericity $\varepsilon = 0.48$, $F(3.80, 380.01) = 35.99$, $p < 0.001$, $\eta^2 = 0.27$. This indicates that the temperatures recorded at different temperature levels differed according to their time point. Contrasts compared each level of temperature to the maximum level across each time point, compared to the 60 minute point and are shown graphically in Figure 39. The first contrast revealed a strongly significant

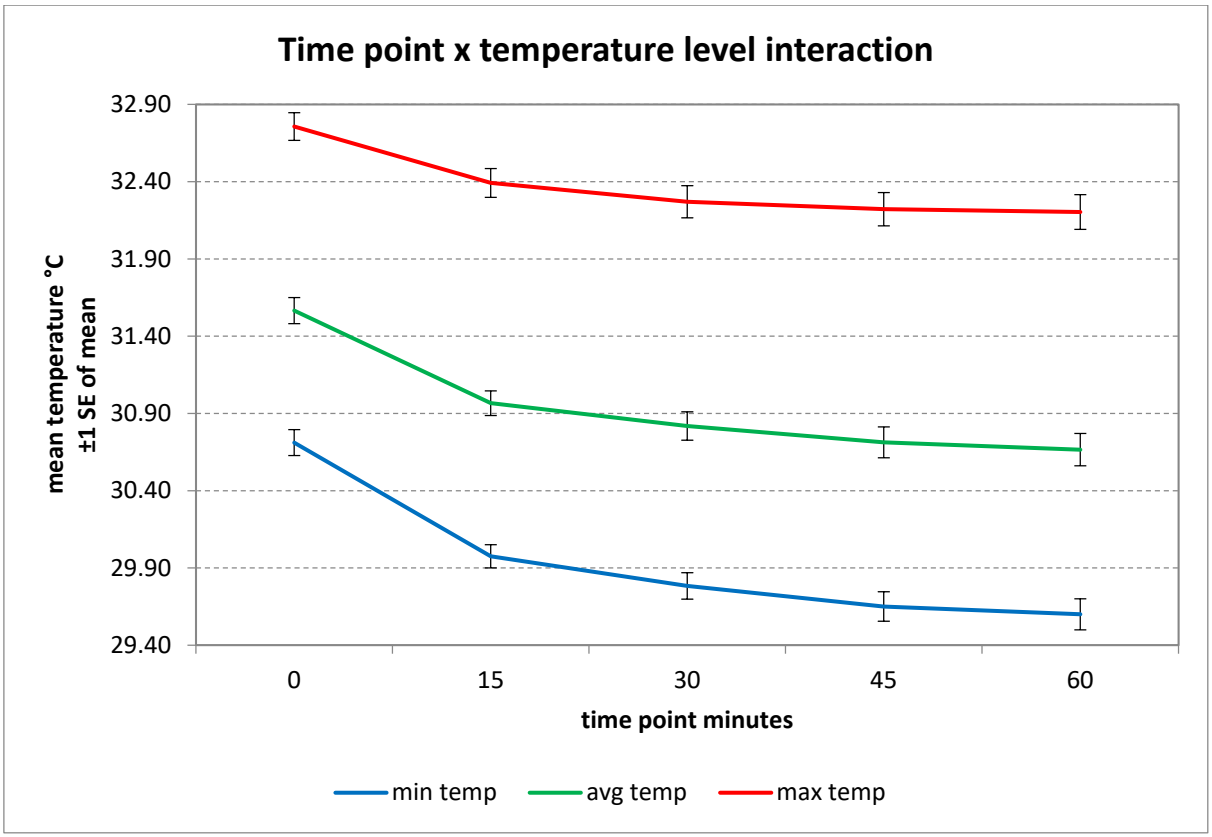


Figure 39. Time x temperature level interaction

interaction when comparing the 0 minutes to the 60 minutes time point, with the minimum temperature level compared to the maximum, $F(1, 100) = 75.47$, $p < 0.001$, $\eta^2 = 0.43$. This shows that with time as the patient acclimatised, there was a greater decline in temperature recorded when the temperature level measured was minimum compared to the maximum level. All other level contrasts were significant except for the last two, which compared the 45 minute to the 60 minute time points, when the minimum temperature level is compared to the maximum, $F(1, 100) = 1.01$, $p = .32$, $\eta^2 = 0.01$ and when the average temperature level is compared to the maximum, $F(1, 100) = 1.64$, $p = .20$, $\eta^2 = 0.02$. This result suggests that beyond 45 minutes, the temperature drop with time were small, and not significantly different between temperature levels. This also suggests that reference to the maximum temperature level will overall provide for a more stable record.

3.3.2 Correlations

Pearson correlations were performed to assess the degree of relationship between temperature levels over the time points and the pool depth. The 15 Pearson correlations are reported in

	pool depth			
temperature level at time point		95% CI		
	r	lower	upper	*N*
t=0 min	.087			114
t=0 avg	.071			114
t=0 max	.077			114
t=15 min	.064			114
t=15 avg	.125			114
t=15 max	.246**	0.066	0.411	114
t=30 min	.061			114
t=30 avg	.114			114
t=30 max	.220*	0.038	0.388	114
t=45 min	.002			114
t=45 avg	.054			114
t=45 max	.144			114
t=60 min	.022			104
t=60 avg	.081			104
t=60 max	.179			104

**. Correlation is significant at the 0.01 level (2-tailed).
*. Correlation is significant at the 0.05 level (2-tailed).

Table 8. Quadrant temperature to pool depth correlation

Table 8. Only two correlations were significant. The maximum temperature level at time point 15 minutes with pool depth was statistically significant, $r = .246$, $p = .008$, 95% CI [0.066, 0.411] and the maximum temperature level at time point 30 minutes with pool depth was also statistically significant, $r = .220$, $p = .019$, 95% CI [0.038, 0.388]. The r^2 was .06 and .05 respectively, indicating that only about 6% and 5% of the variance in pool depth readings could be predicted from the temperature readings at these two levels. This is a weak

	pool depth				
temperature level at time point - wall temperature			95% CI		
	r	*ρ*	lower	upper	*N*
t=0max - wall temp	.252	.012	.066	.421	98
t=15max - wall temp	.423	.000	.267	.571	98
t=30max - wall temp	.420	.000	.257	.569	98
t=45max - wall temp	.384	.000	.212	.538	98
t=60max - wall temp	.403	.000	.224	.558	98
t=0avg - wall temp	.223	.028	.043	.400	98
t=15avg - wall temp	.293	.003	.112	.457	98
t=30avg - wall temp	.332	.001	.166	.487	98
t=45avg - wall temp	.294	.003	.121	.456	98
t=60avg - wall temp	.311	.002	.137	.474	98
t=0min - wall temp	.233	.021	.050	.414	98
t=15min - wall temp	.239	.018	.065	.404	98
t=30min - wall temp	.289	.004	.118	.449	98
t=45min - wall temp	.256	.011	.076	.423	98
t=60min - wall temp	.268	.008	.095	.431	98

Table 9. Δ temperature between quadrant and wall to pool depth correlation

positive relationship showing a tendency for higher thermographic maximum level temperatures to be associated with deeper pool depth.

A second bivariate correlation analysis was carried out, this time using the difference between the temperature levels at time points and the room wall temperature as a computed predictor variable rather than using raw absolute temperature values. This variable was

introduced to control for the effect of the level of ambient room temperature which is a salient factor in thermographic imaging. The results are shown in Table 9. To compute the Pearson correlations a bootstrap estimation was employed in SPSS which is considered a useful robust method for reducing bias in data to realise more precise population estimates. The percentile bootstrap confidence interval shown in the table is drawn from the actual data and will be accurate even if the sampling distribution of *r* is not normal (Wright, London, & Field, 2011). Correlations of temperature levels with pool depth are now all significant and show small to medium positive relationships. The highest correlations were obtained for maximum temperature at time point 15 mins, $r = .423$, $p < .001$, 95% CI [0.267, 0.571] and maximum temperature at time point 30 mins, $r = .420$, $p < .001$, 95% CI [0.257, 0.569]. With a coefficient of determination $r^2 = .18$ at both these levels, the thermographic data can explain 18% of the variance in pool depth readings. This is a medium positive relationship confirming the trend for higher thermographic temperatures to be related with deeper pool depth. This result is shown graphically in Figure 40 where it can be seen that post acclimatisation, the maximum level temperatures show the strongest association to pool depth.

3.3.3 Influence of bio-medical parameters on temperature readings

The next analysis was carried out to assess if the bio-medical parameters recorded have any influence on the relationship between thermographic temperature records and pool depth. Conceptually, we used a moderation model (Field, 2018) to statistically assess any interaction between the predictor (temperature measure) and moderation variables (bio-medical factors) that effects the outcome (pool depth measure). The statistical tool of choice used was the PROCESSv3.5 (Hayes, 2017) plug-in for SPSS which is a widely applied modelling tool in health sciences for assessing effects of interactions in moderation models. The tool can be used with moderators that are continuous, i.e. BMI, Mean abdominal thickness, Systolic, Diastolic blood pressures, pulse, maternal age, gestational age and foetal

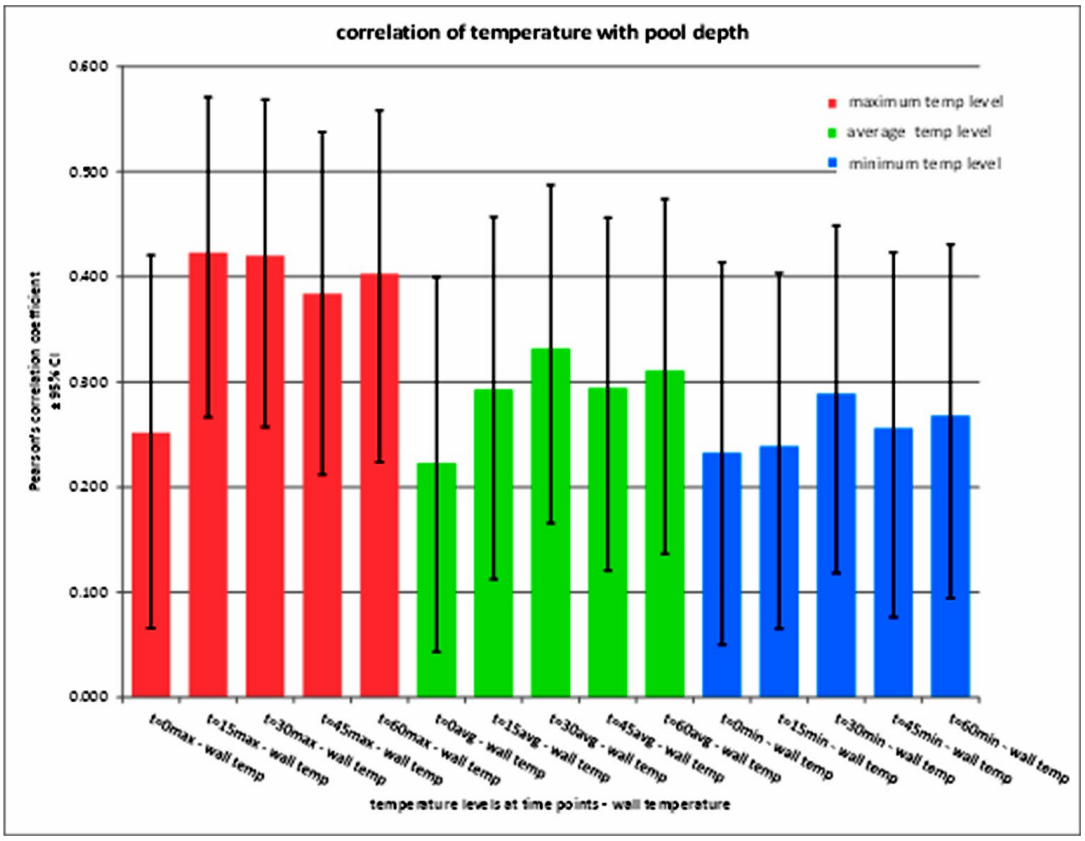


Figure 40. Δ temperature between quadrant and wall to pool depth correlation

abdominal circumference measurements as well as with categorical moderators such as skin type, cream application, placental location, lie and striation. To test for moderation, only the maximum thermographic – wall temperature difference at 15 minutes (QWTt15max) was used as the predictor variable as from the previous results this always reflected the strongest effects. A statistically significant interaction of temperature and moderator in predicting pool depth would indicate a significant moderator influence.

All of these factors except for 'cream applied' were found to have non-significant interactions with temperature in predicting pool depth; (BMI, b = .014, $t(103)$ = -4.99, p = .62; skin type, b = .16, $t(108)$ = .20, p = .84; systolic BP, b = .01, $t(106)$ = .91, p = .36; diastolic BP, b = .01, $t(106)$ = .60, p = .55; pulse, b = .01, $t(106)$ = 1.25, p = .22; foetal lie, b = -1.76, $t(104)$ = -1.44, p = .15; placental location , b = -.08, $t(108)$ = -.35, p = .73; striation, b = .17, $t(106)$ = -.71, p = .48; abdominal thickness, b = .80, $t(93)$ = 1.67, p = .10; maternal age, b = .02, $t(110)$ = .99, p = .32; gestational age, b = .12, $t(110)$ = 1.49, p = .13; abdominal

circumference, $b = .0$, $t(110) = .02$, $p = .98$). For ‘cream application’ which is a dichotomous variable, the overall model was significant, $F(3, 110) = 10.44$, $p < .001$, $R^2 = .22$. The main effect of QWTt15max, $b = 1.21$, $t(110) = 3.88$, $p < .001$ indicates the factor is predictive and as it increases, so does pool depth. This agrees with the correlation analysis discussed above. There was no significant main effect for the cream factor, $b = .14$, $t(110) = .49$, $p = .63$, showing no difference in pool depth means for cream applied and not applied patients. However, the interaction of cream by QWTt15max was significant, $b = -.57$, $t(110) = -2.37$, $p = .02$ and contributed to an R^2 increase of .04. Looking at the conditional interaction effects of QWTt15max at the two values of this moderation, results for ‘no cream’, b = .64, $t(110) = 5.57$, $p < .001$ show that when no cream has been applied, QWTt15max predicts a significantly increased pool depth of 0.64 points, while results for ‘cream applied’, $b = .07$, $t(110) = .34$, $p = .74$ were not significant, indicating that when cream has been applied, QWTt15max is not predictive. Graphing the simple slopes illustrates this in Figure 41.

When cream has been applied there is a non-significant and small positive relationship between QWTt15max and pool depth however with no cream this relationship is still positive but now significant and stronger.

The results of this section indicate that 'cream application' seems to be the only bio-medical factor that interacts with the temperature measure to affect the predicted pool depth. However, although this is effective in the 'no-cream' condition, when cream has been applied thermographic readings are no longer significant predictors of pool depth.

3.3.4 Regression model

The final statistical analysis carried out is a linear regression to suggest the best linear model based on the available data that can serve to describe and qualify the relationship between variables and help predict outcome values based on independent measures. The actual set of predictor variables used in the regression model must be determined by analysis of the data (Riffenburgh, 2006). From the analyses executed so far, the sample data suggests that:

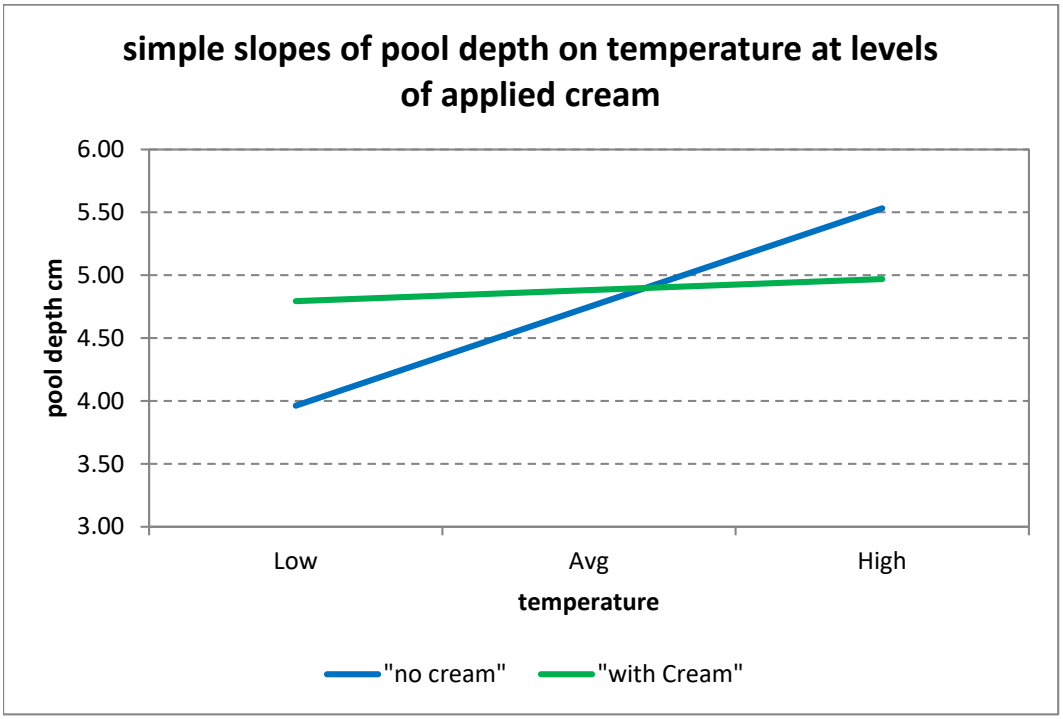


Figure 41. Simple slopes, cream application

i. acclimatisation occurs after 15 to 30 minutes and that the maximum level thermographic readings after this point correlate most strongly with pool depth,
ii. referencing the recorded temperature at the ROI to the wall temperature results in improved correlation with pool depth and
iii. for cases where cream has been applied there is no significant correlation of temperature to pool depth.

Based on these findings, and with the objective of finding a model that balances simplicity and best fit, a backward stepwise multiple regression was performed with pool depth as the dependent variable and maximum temperatures less wall temperature at 15, 30, 45 and 60 minutes as the independent variables. Data of 8 participants with cream applied was removed from the dataset so as not to bias the outcome.

The backward regression resulted in 4 steps with the final step retaining one independent variable that was the maximum temperature less wall temperature at 30 minutes (QWTt30max). Between step one and step four there was no significant change in R^2 (step 1 $R^2 = 0.346$ to step 4, $R^2 = 0.324$, $p = .176$) however although all 4 steps indicated that the relative models were significantly better at predicting the outcome than using no predictors, the model F statistic improved from step 1, $F(4, 63) = 8.323$, $p < .001$ to step 4, $F(1, 66) = 31.648$, $p < .001$. This result can be interpreted as meaning that the final model is markedly better at predicting the outcome variable. It is noted that the variation inflation factor (VIF) for the models in steps 1 to 3 was greater than 10 which is taken to suggest potential multicollinearity between the variables (Hahs-Vaughn, 2016). This is not unexpected since these temperature time-point variables are within subjects, however now a simple linear regression with just the QWTt30max variable as the sole predictor was carried out to ensure regression assumptions are met.

The results of this simple linear regression suggest that a significant proportion of the total variation in pool depth was predicted by QWTt30max, F (1, 80) = 31.136, $p < .001$, $R^2 = .28$. This indicates that this model can account for 28% of the variation in pool depth. Additionally, for QWTt30max the unstandardized slope (.575) and standardized slope (.529) are statistically significantly different from zero ($t(80) = 5.580$, $p < .001$); with every one point increase in temperature difference, pool depth will increase by approximately 1/2 of one point. The confidence interval around the unstandardized slope does not include zero, 95% CI [0.370, 0.780] further confirming that QWTt30max is a statistically significant predictor of pool depth. The resulting predicative model is

$$pool\ depth\ = -1.117 + 0.575\ \times QWTt30max$$

Figure 42 illustrates this relationship graphically giving an indication of confidence and prediction intervals. The CI indicates the limits within which there is a 95% probability the true population regression line from the sample data lies. The prediction interval (PI) marks the bounds around the predicted value of pool depth wherein for a specific value of temperature there is a 95% probability the true value of pool depth lies.

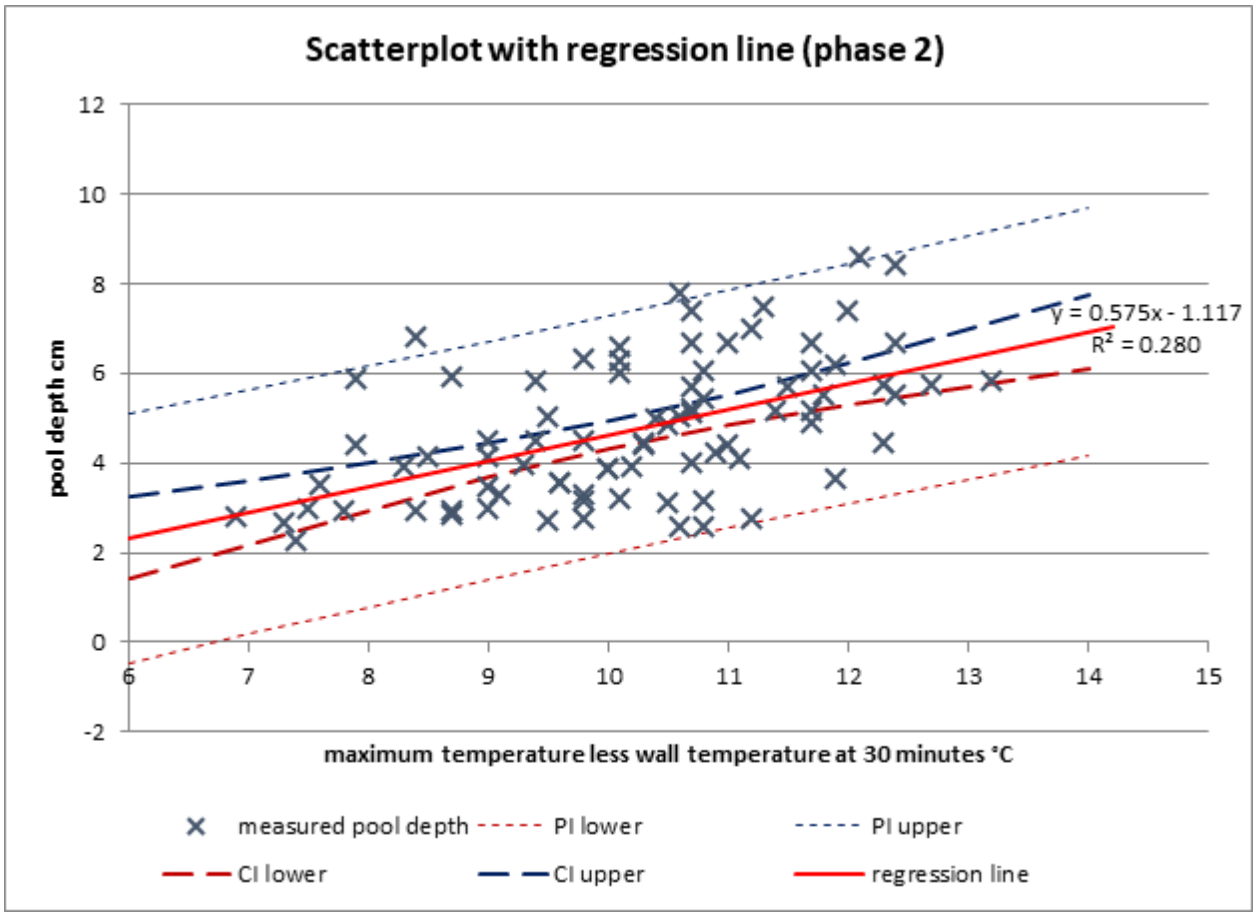


Figure 42. Regression plot, Phase 2

3.4 Phase 3 – Overview

The goal of Phase 3 of the study was to investigate if the relationship between thermographic temperature and the maximum liquor pool depth in healthy subjects suggested in Phase 2 would extend to patients diagnosed with any form of diabetes. As described in the methodology, availability of participants was limited because of the reduced incidence of this condition for the cohort profile defined for the study. For this reason, 10 patients were recruited, resulting in the number of raw data collected yielding $N = 40$ for this sample.

The sections below describe the rationale and results of the statistical analyses undertaken. Prior to this however, the descriptive statistics of the demographic and biological measurements is presented.

3.4.1 Demographic and Biological Measurements

Out of the 10 patients that participated in the study, three patients suffered from IDDM (Insulin Dependent Diabetes Mellitus), one suffered from NIDDM (non-Insulin Dependent Diabetes Mellitus) and six suffered from GDM (Gestational Diabetes Mellitus).

3.4.1.1 Maternal age and Gestational age

Data in Table 10 shows that the average maternal age was 31 years, with the youngest patient being 20 years of age and the eldest age being 41 years. The gestational age of the patients ranged from 24 weeks and 2 days, to 33 weeks and six days. The mean gestational age was 29 weeks.

Patient Number	Maternal Age	Gestational Age
1	36	33+4
2	41	27+4
3	27	33+6
4	29	25+3
5	20	27+5
6	32	32+2
7	31	24+2
8	28	25+4
9	33	29+2
10	37	29+1
Mean	**31**	**29**
SD	**5.79**	**3+6**

Table 10. Maternal and gestational age, Phase 3

3.4.1.2 Skin Type

The skin type was quantified by using the Fitzpartick Phototype scoring. 7 patients had a Type 3 skin score whilst 3 patients had a Type 4 skin score.

3.4.2 Abdominal Features

3.4.2.1 Abdominal Striations

Out of the 10 patients that participated in the study, five had visible striations and 5 did not.

3.4.2.2 Use of Cream

Out of the 10 patients that participated, three patients used cream regularly and did so on the day of the study all three applied the cream more than one hour prior to the start of the study.

Patient Number	Systolic BP	Diastolic BP	Pulse
1	114	68	83
2	143	90	85
3	127	77	85
4	138	88	91
5	133	58	93
6	144	82	77
7	115	74	84
8	120	67	69
9	118	76	61
10	105	65	89
Mean	**126**	**75**	**82**
SD	**12.7**	**9.74**	**9.53**

Table 11. Blood pressure, pulse - Phase 3

3.4.3 Biological and Clinical Parameters

3.4.3.1 Use of Diabetic Medication

This sub-group of patients suffered from diabetes in pregnancy. These patients were on medications as treatment for this condition. These medications belonged to two groups; Oral Hypoglycaemic Agents (Metformin® being the most common agent)(OHA), and Insulin (both long acting such as Insulitard® and short acting such as Humilin®). One patient was on no medication and was on dietary advice alone. Six patients were on OHA's and three patients were on Insulin regimens.

3.4.3.2 Blood Pressure and Pulse

Each patient had their blood pressure and pulse measured using a digital monitor during the experiment. Table 11 shows the individual readings. The mean systolic blood pressure was 126 mmHg, the mean diastolic blood pressure was 75 mmHg and the mean pulse was 82 bpm.

3.4.3.3 Temperature

The Oral temperature for each patient was taken using a digital thermometer. This is compared to the temperature in the room, as extracted from the wall temperature on the thermographic image of that same patient. This is also compared to the relative temperature that day. The graph in Figure 43 clearly shows that the oral temperature remains stable from

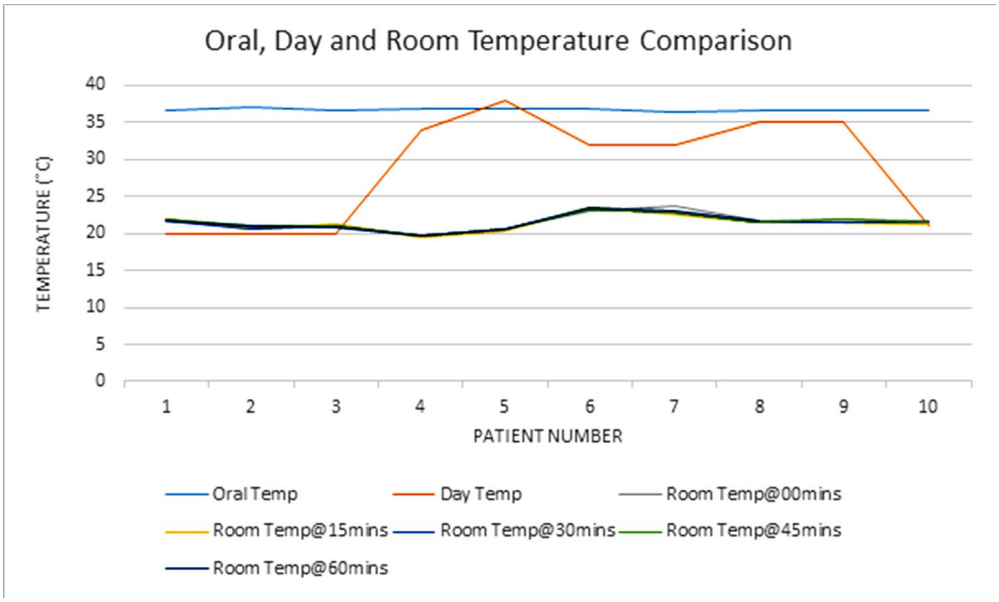


Figure 43. Temperatures, Phase 3

patient to patient, whilst the outside day temperature does not. This is expected as the core temperature of the human body is not expected to fluctuate greatly in response to larger ambient changes, rather it should remain as stable as possible despite these fluctuations in order to maintain stable bodily function.

3.4.4 Ultrasound Data

3.4.4.1 Liquor Pool Depth

For each patient, four quadrants were measured by ultrasound in order to obtain the depth of liquor in each quadrant. These measurements were carried out from right lower to left lower quadrant in a clockwise fashion. The pool depths are shown in Figure 44, with the mean pool depth measuring 4.94cm.

The minimum pool depth was 1.16cm and the maximum pool depth was 10.49cm. This variation is expected when measuring liquor volume. It is the addition of all four quadrants in the AFI (Amniotic Fluid Index) or the measurement of the deepest pool, which is of clinical significance, meaning that the individual values on their own may show a large variation between them, even within the same patient.

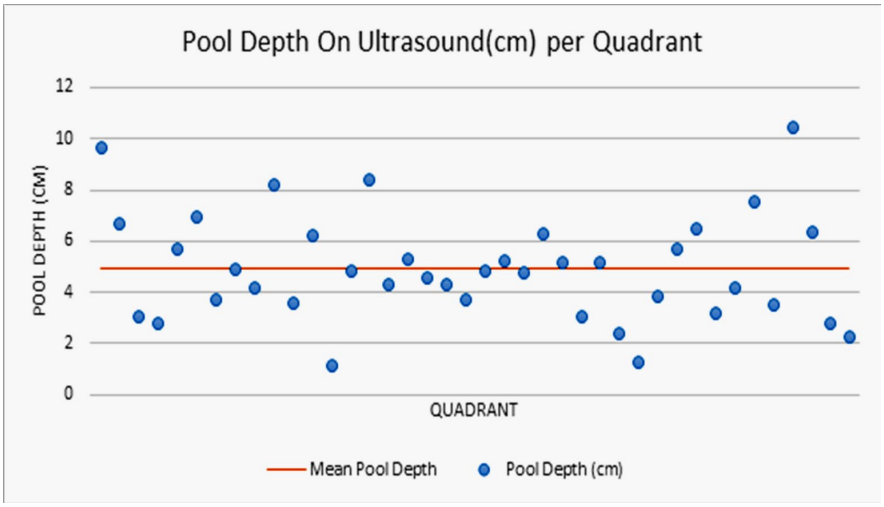


Figure 44. Pool depth scatter plot, Phase 3

3.4.4.2 Placental Location

Out of the 10 ultrasound scans carried out, the placenta was found to be Fundal in two patients, Posterior in five patients and Anterior in three patients.

3.4.4.3 Foetal Lie

Out of the 10 patients assessed during the study, one foetus was found to be transverse lie, two were found to be in breech position and seven were found to be in cephalic position.

3.4.4.4 Mean abdominal thickness

The mean abdominal thickness was measured by measuring the thickness around the

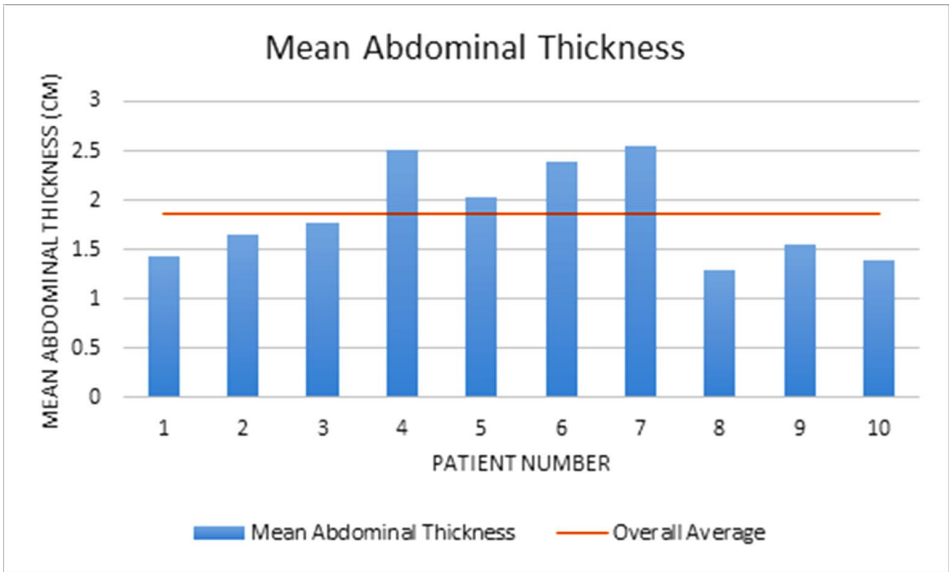


Figure 45. Mean abdominal thickness per patient, Phase 3 *(SD=0.46)*

umbilicus for each patient, shown in Figure 45. The measurement consisted of the thickness of the umbilicus, followed by that at 6 o'clock, 9 o'clock, 12 o'clock and 3 o'clock. These were carried out using a linear probe on the ultrasound. The overall mean abdominal thickness was 1.91cm, with the thicknesses ranging between 1.3cm and 2.56cm.

3.5 Phase 3 - Data screening

As for Phase 2, the new data set was screened for missing data, accuracy and consistency to identify and resolve possible issues before undertaking the data analyses. As the sample size in this phase was relatively small, special attention was given to assess the assumptions of normality, linearity and homoscedasticity are fulfilled. Outliers were initially screened by reviewing box-plots for each of the predictor variables. Pool depth results were reviewed for reasonableness and accuracy given that the subjects in this phase had no

indicated obstetric complications. Finally, in view of the results obtained in Phase 2, data for participants who were recorded as having 'cream applied' was filtered out of the analyses.

The net screened dataset for this diabetic patient group resulting after appropriate screening and filtering as utilized in the analyses is N = 23, $M_{\text{pool depth}}$ = 4.76 cm, $SD_{\text{pool depth}}$ = 1.90, $Range_{\text{pool depth}}$ = 1.29 – 8.42 cm. As per the sample size to power graph shown in Figure 30 in the Methodology chapter, this results in a power estimation of 82% which is deemed acceptable (Cohen, 1992) for an eventual regression analysis.

Formal statistical tests of normality were used to gauge for each variable the degree to which the sample distribution differs from a normal distribution. This approach was used in view of the relatively small sample size. The Shapiro-Wilk test was run using SPSS as it is generally deemed a powerful test for normality and is suggested for small sample sizes with N less than 50 (Hahs-Vaughn, 2016). Non-statistically significant outcomes imply that the distribution of the variable's sample is not significantly different from a normal distribution. This test indicated that 5 of the variables did not follow the normal distribution (Temperature Quadrant t=0 min, p = .040; Temperature Quadrant t=0 avg, p = .004; QuadWallTt15min, p = .013; QuadWallTt0avg p = .002; QuadWallTt60max, p = .015) and any inferences using these variables are considered with caution.

3.6 Phase 3 - Statistical results

As for Phase 2, an initial analysis of the thermographic data was done to find if any significant effects of time and ROI are evident. This was followed by a correlation analysis between the thermographic data and pool depth. Results are reported below.

3.6.1 Significance of group differences

The effects on thermographic data over time and location were explored by conducting a 3x5x4 mixed ANOVA as for Phase 2. Using Greenhouse-Geisser correction for sphericity there were significant main effects of temperature level and time point and their interaction. With Greenhouse-Geisser correction for sphericity, significant main effects of

temperature level, time point and their interaction were again indicated. The main effect of temperature level was significant, $F(1.05, 15.79) = 204.21$, $p < 0.001$, $\eta^2 = 0.93$, $\varepsilon = 0.53$ which means that if all other factors are ignored, the mean temperature recorded was significantly different between temperature levels. Also the main effect for time point was also significant, $F(1.19, 17.90) = 17.81$, $p < 0.001$, $\eta^2 = 0.54$, $\varepsilon = 0.30$ which means that disregarding other factors, the mean temperature recorded was significantly different between time points. The interaction between temperature and time points was significant $F(3.05, 45.74) = 6.25$, $p < 0.001$, $\eta^2 = 0.29$, $\varepsilon = 0.38$. Within subject contrasts showed that mean temperatures for 0 minutes - $F(1, 15) = 19.27$, p 0.001, 15 minutes - $F(1, 15) = 42.31$, $p <$ 0.001 and 30 minutes - $F(1, 15) = 17.63$, $p = 0.001$ were significantly higher than that for 60 minute time point. However, the difference between temperatures for the 45 and 60 minute time points was non-significant - $F(1, 15) = 2.02$, $p = .175$. Finally, the main between subjects effect of pool location was not significant $F(3, 15) = .22$, $p = .88$, $\eta^2 = 0.043$ indicating no significant difference between mean temperatures for the 4 quadrants. These results are generally as expected and in line with the general results obtained for the non-diabetic cohort. However, due to the small sample size especially when grouped by quadrants, the non-normality of some variables and a resulting non-homogeneity for Temperature Quadrant t=15max (Levene's Test $p = .003$) the results for these statistical tests are only indicative.

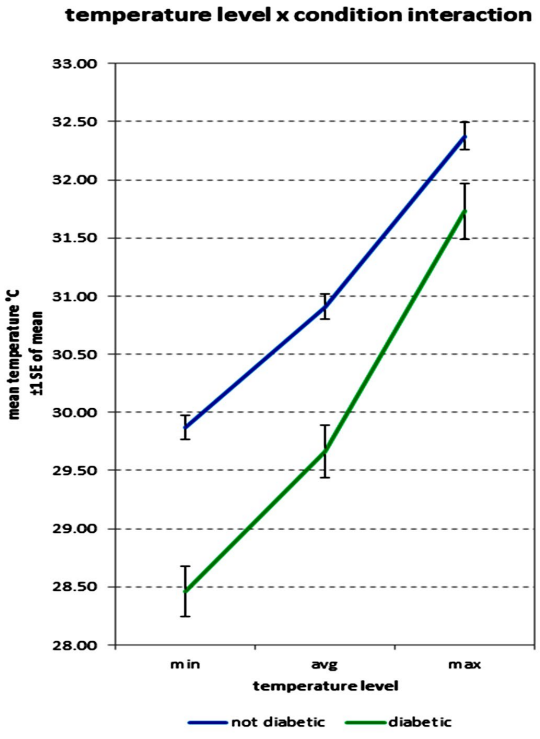


Figure 46. Temp level x clinical condition interaction

More informative is the comparison of the mean temperature over time-points and levels per clinical condition (diabetic or non-

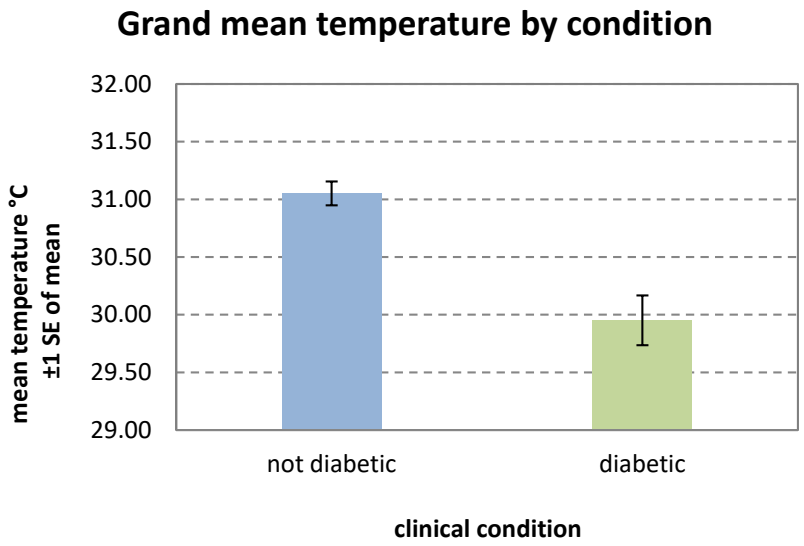


Figure 47. Mean temperature by clinical condition

diabetic) depicted in Figure 47. For non-diabetic patients $M = 31.05$, CI [30.845, 30.997] while for diabetic participants $M = 29.95$, CI [29.523, 30.381], approximately 1°C lower. These means are also significantly different $F(1, 83) = 21.116$, $p < 0.001$, $\eta^2 = 0.203$. There was a significant difference between temperature levels per condition interaction, both in the case of the minimum compared to the maximum level ($F(1, 83) = 16.49$, $p < 0.001$, $\eta^2 = 0.166$) and in the case of the average compared to the maximum level ($F(1, 83) = 22.66$, $p < 0.001$, $\eta^2 = 0.214$). The difference of mean temperature over different levels between clinical groups is illustrated in Figure 46.

3.6.2 Correlations

Pearson bivariate correlation analysis was carried out to assess the degree of relationship between temperature levels over the time points, the difference between the temperature levels at time points and the room wall temperature, and the pool depth. Again the bootstrap estimation was employed in SPSS as a mitigating method for reducing normality bias in the data giving accurate confidence intervals even if the *r* sampling distribution is non-normal (Wright et al., 2011). All correlations for temperature levels over the time points with pool depth proved non-significant and while some temperature

	pool depth				
temperature level at time point - wall temperature			95% CI		
	r	*p*	lower	upper	*N*
t=0max - wall temp	0.125	.285	-.383	.519	23
t=15max - wall temp	0.308	.076	-.181	.688	23
t=30max - wall temp	0.420	.023	-.053	.733	23
t=45max - wall temp	0.443	.017	-.008	.738	23
t=60max - wall temp	0.448	.027	-.060	.746	19
t=0avg - wall temp	0.095	.333	-.444	.514	23
t=15avg - wall temp	0.429	.021	.014	.704	23
t=30avg - wall temp	0.435	.019	-.034	.740	23
t=45avg - wall temp	0.499	.008	.044	.778	23
t=60avg - wall temp	0.521	.011	.031	.784	19
t=0min - wall temp	-0.034	.439	-.517	.408	23
t=15min - wall temp	0.345	.054	-.145	.670	23
t=30min - wall temp	0.315	.072	-.109	.635	23
t=45min - wall temp	0.178	.208	-.305	.572	23
t=60min - wall temp	0.219	.184	-.325	.668	19

Table 12. Δ temp between quadrant and wall to pool depth correlations, Phase 3
(factors marked in blue failed the Shapiro-Wilk normality test)

difference correlations were significant, 3 factors failed normality tests as shown in results Table 12. The same data is shown graphically in Figure 48. By inspection, the most useful results of the correlation analysis are the variables 't=45avg - wall temp', $r = .499$, $p < .008$, 95% CI [0.044, 0.778] and 't=60avg - wall temp', $r = .521$, $p < .011$, 95% CI [0.031, 0.784]. While the correlations of temperature levels with pool depth are significant and show small to medium positive relationships, the wide confidence interval of these point results indicates a greater amount of random error due to the small sample size suggesting the estimated *r*'s are not as precise and inference beyond the sample needs to be treated with caution.

3.6.3 Influence of diabetic condition on temperature readings

Using the combined dataset of diabetic and non-diabetic participants, that is combining the datasets of Phase 2 and Phase 3, a moderation model was evaluated with PROCESSv3.5 (Hayes, 2017). The intent was to statistically assess if any interaction between the predictor (temperature measure) and diabetes condition has any influence on the relationship between thermographic temperature records and pool depth. Following the

correlation results, to test for moderation, the average thermographic less wall temperature difference at 45 minutes (QWTt45avg) and the average thermographic less wall temperature difference at 60 minutes (QWTt60avg) were used as predictor variables in separate moderation tests. The latter factor showed no significant interaction (b = 80, $t(101)$ = 1.73, p = .09), however, for QWTt45avg there was a significant interaction effect with diabetes condition on pool depth; b = 1.04, $t(101)$ = 2.01, p = .047, 95% CI [0.012, 2.065] which contributed to an R^2 increase of .03. Looking at the conditional interaction effects of QWTt45avg at the two values of this moderation, results for 'not diabetic' condition; b = .484, $t(101)$ = 4.01, p < .001, 95% CI [0.245, 0.723] showed, as expected, results in line with those already reported for Phase 2, while results at the 'diabetic' condition, b = 1.523, $t(101)$ = 3.03, p < .05, 95% CI [0.525, 2.521] indicated a steeper influence of temperature on pool depth. Although significant, this effect shows a wide confidence interval suggesting noisy data for this condition group as already indicated in the correlation tests. For both

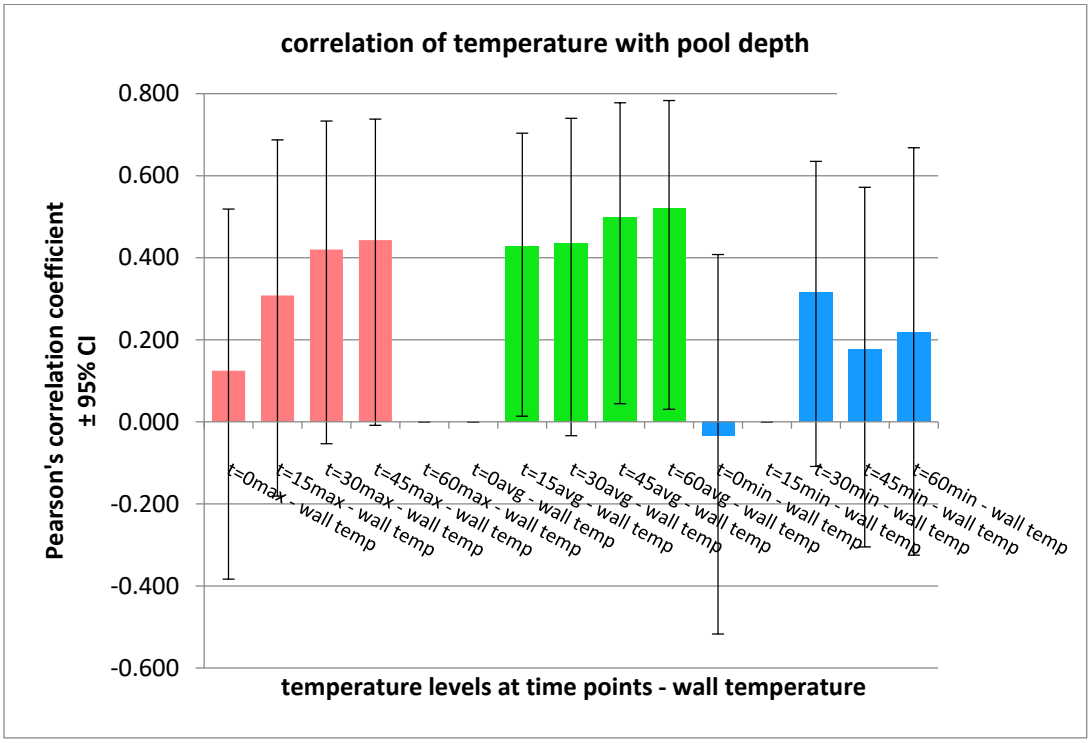


Figure 48. Δ temp between quadrant and wall to pool depth correlations, Phase 3

temperature factors there was no significant main effect of diabetic condition factor, $b = .33$, $t(101) = .909$, $p = .366$ and $b = .26$, $t(101) = .662$, $p = .51$ respectively.

For comparative purposes a moderation analysis of clinical condition on QWTt30max, which was the regression analysis predictor variable used in Phase 2 was also conducted. The results showed that as for the QWTt60avg case, there was no significant interaction of the clinical condition factor ($b = .40$, $t(101) = 1.03$, $p = .31$).

A moderation analysis was carried out to assess the interaction of the temperature measurements and the biomedical factors within this clinical group for QWTt45avg, as this was found to have a significant interaction with the diabetic condition on pool depth. Maternal age, gestational age, skin type, mean abdominal thickness, striations, systolic blood pressure, diastolic blood pressure, pulse, medication-OHA, medication-insulin, placental location and foetal lie were all found to have no significant interaction with QWT45avg on pool depth (skin type, $b = 1.16$, $t(25) = 1.08$, $p = .29$; systolic BP, $b = .01$, $t(25) = .34$, $p = .74$; diastolic BP, $b = -.004$, $t(25) = -.06$, $p = .95$; pulse, $b = -.03$, $t(25) = -.46$, $p = .65$; medication-OHA, $b = .09$, $t(25) = .86$, $p = .40$; medication-insulin, $b = -1.50$, $t(25) = -1.54$, $p = .14$; foetal lie, $b = 2.44$, $t(23) = 1.88$, $p = .07$; placental location , $b = -1.03$, $t(23) = -.96$, $p = .35$; striation, $b = .41$, $t(25) = .43$, $p = .68$; abdominal thickness, $b = -1.78$, $t(23) = -1.56$, $p = .13$; maternal age, $b = -.05$, $t(25) = -.34$, $p = .74$; gestational age, $b = .21$, $t(25) = 1.67$, $p = .11$).

There was however a significant interaction with cream, reflecting the results of Phase 2. The individual effect of the use of cream is non-significant $b = 0.3$, $t\ (25) = 0.38$, $p = 0.71$, 95% CI [-1.3, 1.89. The interaction of the use of cream and QWTt45avg on the pool depth however, was found to be significant - $b = -2.8$, $t\ (25) = -3.3$, $p = 0.003$, 95% CI [-4.55, -1.05]. The conditional effect of not using cream was $b = 1.52$, $t\ (25) = 2.78$, $p = 0.01$, 95% CI [0.39, 2.65] and that of using cream was $b = -1.28$, $t\ (25) = -1.97$, $p = 0.06$, 95% CI [-2.61, 0.06].

Another significant interaction was that of the abdominal circumference (AC) measurements on ultrasound and QWT45avg. The individual effect of the AC was not significant $b = 0.005$, $t\ (21) = 0.48$, $p = 0.64$, 95% CI [-0.02, 0.03], however the interaction with QWTt45avg on pool depth was significant; $b = 0.023$, $t\ (21) = 2.23$, $p = 0.04$, 95%CI [0.002, 0.04]. For measurements 1SD above the mean AC, the conditional effect of this predictor are significant $b = 1.74$, $t\ (21) = 2.28$, $p = 0.03$, 95% CI [0.15,3.33]. With a measurement of at least 29.2cm, pool depth and QWTt45avg are significantly related $b = 1.27$, $t\ (21) = 2.08$, $p = 0.05$, 95% CI [0.00, 2.55]. This relationship becomes more significant as the AC measurement increases, with the highest at 31.5 $b = 1.81$, $t\ (21) = 2.3$, $p = 0.03$, 95% CI [0.17, 3.47].

There was no statistically significant interaction with gestational age.

With regards to the effect of diabetic medication taken by these patients, there was no significant interaction with OHA or Insulin and QWT45avg on pool depth $b = 0.88$, $t\ (25) = 0.86$, $p = 0.4$, 95% CI [-1.23, 2.99] and $b = -1.5$, $t\ (25) = -1.54$, $p = 0.14$, 95% CI [-3.4, 0.5] respectively.

3.6.4 Regression model

A simple linear regression analysis was carried out to suggest the linear model based on the available data that can best depict the relationship between thermographic readings and pool depth for the diabetic cohort. Previous statistical results for Phase 3 data suggest a simple linear regression with just the QWTt45avg factor as the predictor variable would be the most appropriate. Since the sample is small, the bootstrapped confidence intervals for the regression coefficient and significance value were also evaluated because they do not rely on assumptions of normality or homoscedasticity.

The linear regression indicates that QWTt45avg can explain a portion of the total variation in pool depth with statistical significance, $F\ (1, 21) = 6.97$, $p = .015$, $adjR^2 = .21$. The adjusted R^2 statistic is being reported here because when a small sample is involved it

adjusts for the tendency of R^2 to overestimate the value (Tabachnick & Fidell, 2013). The result indicates that this model can account for 21% of the variation in pool depth. Additionally, the unstandardized slope is statistically significantly different from zero, β = 1.523, $t(21) = 2.64$, $p = .015$. The bootstrapped confidence interval and significance around the unstandardized slope, 95% CI [.133, 2.708], $p = .024$ so even when not relying on the assumptions of normality or homoscedasticity, QWTt45avg is still confirmed as having a positive relationship with pool depth. The wide CI and increased bootstrapped $SE = .65$ however, again reflect the noisy data in the sample. The resulting regression model is

$$pool\ depth\ =\ -8.049 + 1.523\ \times QWTt45avg$$

Figure 49 illustrates this relationship graphically giving an indication of confidence and prediction intervals of the regression line.

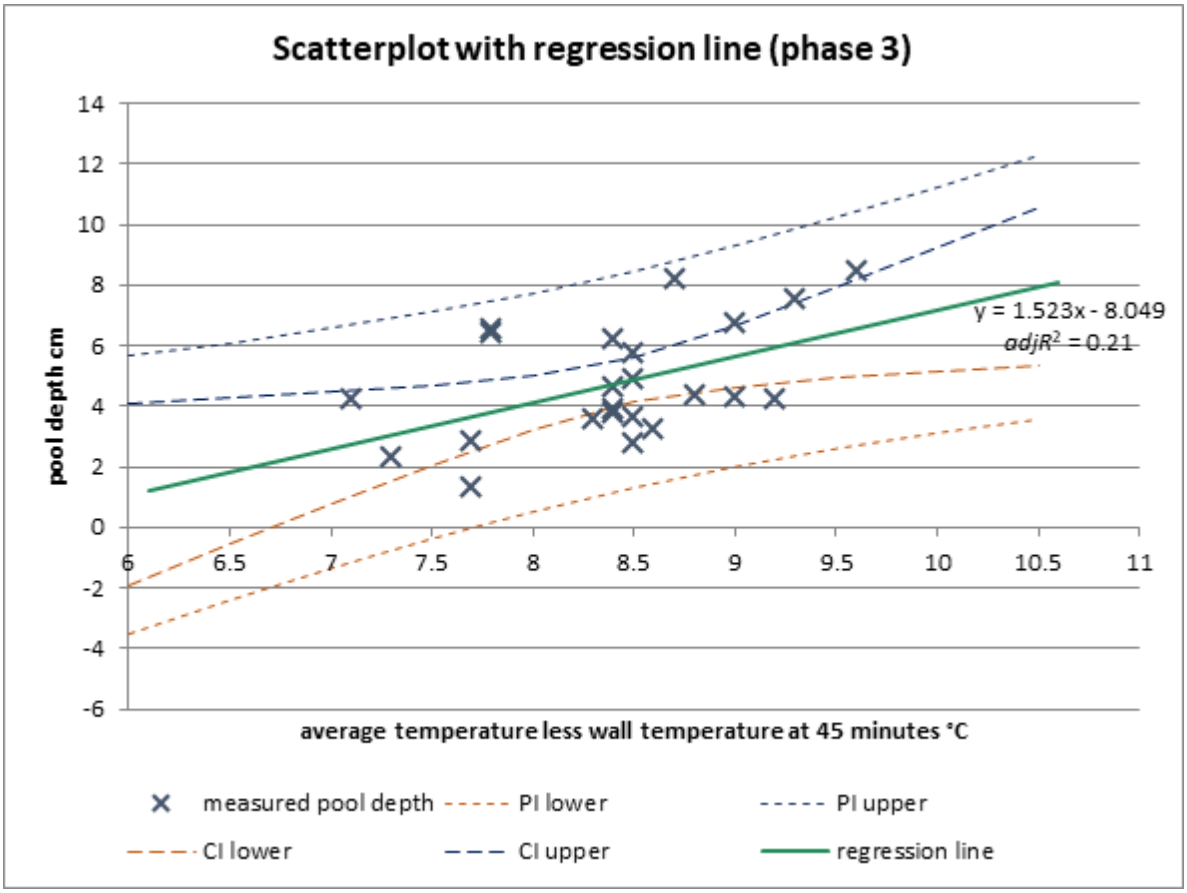


Figure 49. Regression line, Phase 3

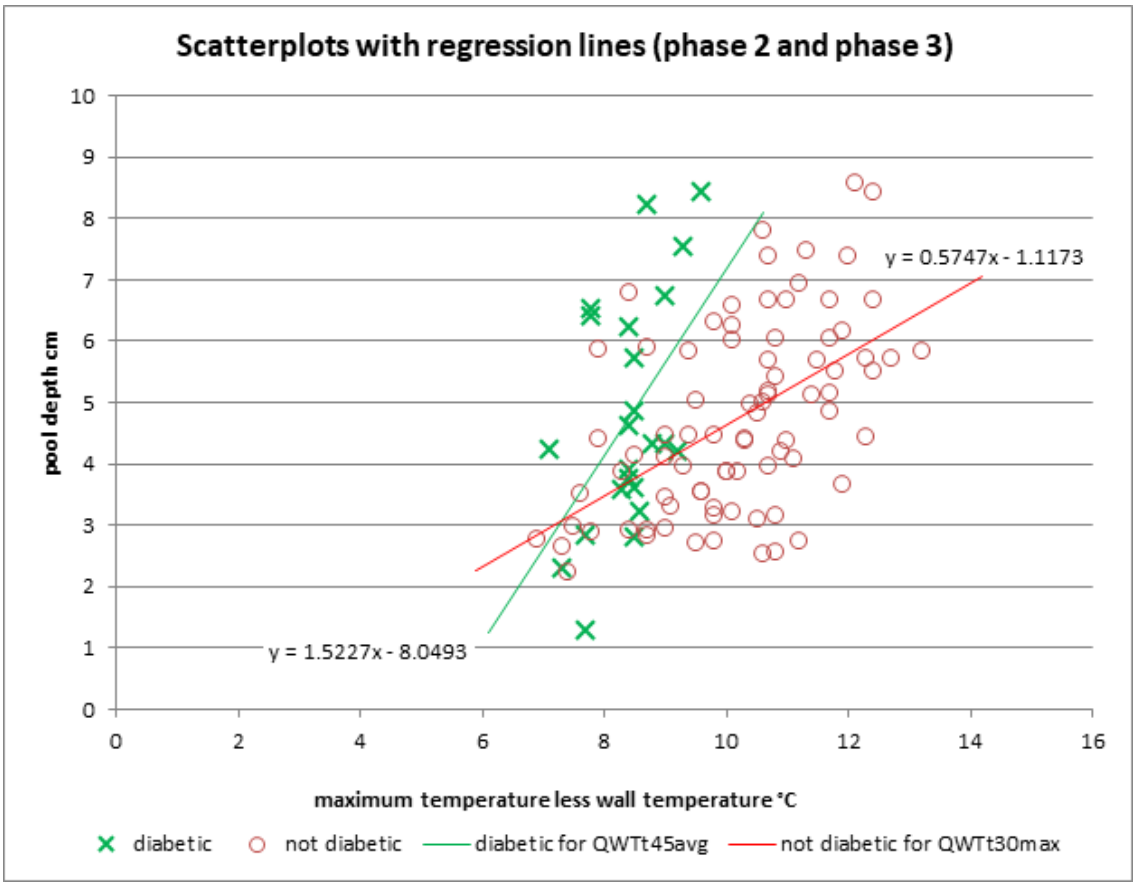


Figure 50. Comparative regression lines Phase 2 / Phase 3

The regression lines are compared in the grouped scatter plot shown in Figure 50. It is to be noted that the temperature values for the two conditions are taken at different time-points and temperature levels. Considering the combined datasets, an independent t-test of mean predicted pool depth for diabetic and non-diabetic participants yields a non-significant difference of means t(103) = -.062, p = .95 which indicates that the clinical condition has no significant effect on predicted pool depth from thermographic data. This was also confirmed by the moderation results above, where clinical condition was a non-significant predictor at the selected temperature values. Given that the mean temperature difference for QWTt45avg over the diabetic sample data is M = 8.41, 95% CI[8.14, 8.68] and for QWTt30max, non-diabetic sample M = 10.21, 95% CI[9.89, 10.52] and allowing for this difference in mean temperature, the regression lines would deliver similar pool depth predictions within the CI's of each line.

3.7 Reliability – Results

3.7.1 Intra-rater Reliability Test

In total there were 60 data sets for each operator, 15 thermographic images with 4 ROI measurements each. For each ROI, the minimum, maximum and average temperatures were analysed. By using SPSS, the mean, standard deviation (SD), ICC and the 95% Confidence Interval of the ICC were calculated as seen in Table 13.

The mean temperature of the minimum temperature level readings within the ROI's of operator 1 was 32.01˚C with a standard deviation of 1.35. The mean temperature of the minimum temperature readings within the ROI's for operator 2 was 31.54˚C with a standard deviation of 1.51. The mean temperature of the maximum temperature within the ROI's for operator 1 was 34.93˚C with a standard deviation of 1.32, and for operator 2 was 34.29˚C with a standard deviation of 1.71. The mean temperature for the average temperature within

	Mean	**SD*[1]**	**ICC*[2]**	**95% CI*[3]**	**Cronbach's α**
Min Operator 1	32.01	1.3449	0.875	0.721-0.936	0.899
vs Min Operator 2	31.54	1.5057			
Max Operator 1	34.93	1.3201	0.831	0.595-0.917	0.870
vs Max Operator 2	34.29	1.7145			
Average Operator 1	33.25	1.2965	0.868	0.673-0.983	0.90
vs Average Operator 2	32.72	1.5918			

Table 13. Intra-rater Reliability test results

*[1] SD = Standard Deviation
*[2] ICC = Intraclass Correlation Coefficient
*[3] 95% CI = 95% Confidence Interval

the ROI's for operator 1 was 33.25˚C with a standard deviation of 1.30, and for operator 2

was 32.72˚C with a standard deviation of 1.60. The standard deviations are rounded up to two decimal places in the description of the table above.

There was excellent level of agreement between the operators for minimum, maximum and average temperatures within the ROI's (ICC – 0.875, 0.831 and 0.868 respectively). The Cronbach's alpha was 0.899 for Minimum temperature readings, 0.870 for Maximum temperature readings and 0.90 for Average temperature readings, showing high internal consistency for all three test comparisons.

3.7.2 Test-Retest Reliability

In total there were 60 data sets measured by the operator. The same 15 thermographic images were analysed 4 days apart, under the same conditions in the same setting. During the analysis, the operator demarcated 4 ROI and for each, the minimum, maximum and average temperatures were collected. These readings were collected manually and transferred onto an Excel sheet. They were arranged in an order to compare the two consecutive tests for minimum, maximum and average temperature respectively.

By using SPSS, the mean, standard deviation (SD), ICC and the 95% Confidence Interval of the ICC were calculated as seen below. The results were as follows.

The mean temperature for the minimum temperature readings within the ROI's during test 1 was 29.91˚C with a standard deviation of 1.29. The mean temperature of the minimum temperature readings during test 2 was 29.99˚C with a standard deviation of 1.70. The mean temperature of the maximum readings during test 1 was 33.00˚C with a standard deviation of 1.35 and that during test 2 was 33.03˚C, with a standard deviation of 1.29. The mean temperature of the average readings during test 1 was 31.1˚C, with a standard deviation of 1.23, and during test 2 was 31.09˚C with a standard deviation of 1.19.

There was excellent level of agreement between the repeat tests of the maximum temperature and average temperature readings within the ROI's (ICC – 0.989 and 0.997

respectively. There was however a moderate level of agreement between the repeat tests of the minimum temperature readings within the ROI's (ICC – 0.689).

In Table 14, one can see that there is a close agreement between the MDC_{95} values of the Maximum temperature readings (0.484 and 0.462) and Average temperature readings (0.439 and 0.427). There is however a noticeable difference between the MDC_{95} values of the Minimum temperature (0.462 and 0.608).

	Mean	**95% CI*[1]**	**SD*[2]**	**SEM*[3]**	**MDC_{95}*[4]**
Min test 1	29.91	30.24 – 29.58	1.2938	0.1670	0.462
vs Min test 2	29.99	30.42 – 29.56	1.7015	0.2196	0.608
Max test 1	33.00	33.35 – 32.66	1.3528	0.1746	0.484
vs Max test 2	33.03	33.35 – 32.70	1.2937	0.1670	0.462
Average test 1	31.10	31.41 – 30.79	1.2271	0.1584	0.439
vs Average test 2	31.09	31.39 – 30.78	1.1944	0.1541	0.427

Table 14. Test Retest reliability results showing individual test statistics

*[1] 95% CI = 95% Confidence Interval; *[2] SD = Standard Deviation; *[3] SEM = Standard Error of the Mean; *[4] MDC_{95} = Minimal Detectable Change at 95% Confidence Interval

In Table 15, one can also note the similarity in MDC_{95} between the Maximum temperature group and the Average temperature group, as compared to the Minimum temperature group (0.38, 0.18 and 2.31 respectively). This is reflected in the SEM values for the same (0.138, 0.066 and 0.835). This shows that the reliability of the maximum temperature and average temperature readings within the ROI on thermogram is much higher than the minimum temperature readings.

	ICC*[5]	95% CI	Cronbach's α	Mean SD	SEM	MDC_{95}
Min test 1 vs Min test 2	0.689	0.479 – 0.815	0.686	1.479	0.835	2.31
Max test 1 vs Max test 2	0.989	0.982 – 0.994	0.989	1.323	0.138	0.38
Average test 1 vs Average test 2	0.997	0.995-0.998	0.997	1.210	0.066	0.18

Table 15. Test Retest reliability results showing comparative test statistics

*[5] ICC = Interclass Correlation Coefficient

3.8 Summary of results

The results of the pilot study found a non-significant correlation between the deepest liquor pool depth measured on ultrasound and the thermographic temperature measured at that same pool (r = -.24, p = .099, 95% CI [-.543, .077]). However, this provided the basis for further analysis by using a refined methodology in the subsequent main studies. The study with the healthy pregnant group, showed that there was there was a significant positive correlation between the liquor pool depth measurement and the maximum temperature measured by thermography at 30 minutes (r = .220, p = .019, 95% CI [0.038, 0.388]). This relationship was found to be even stronger when the wall temperature, which showed a stable ambient temperature over time, was subtracted from the absolute temperatures (r = .420, p < .001, 95% CI [0.257, 0.569]). In the diabetic group, this was found to be at 45 minutes when the wall temperature was subtracted from the average temperature (r = .499, p < .008, 95% CI [0.044, 0.778]). The mean temperature over time between different levels in the healthy group was also shown to be significant (F (3.80, 380.01) = 35.99, p < .001, η^2 = 0.27), as was the case in the diabetic group (F (1.05, 15.79) = 204.21, p < .001, η^2 = 0.93). The conditional

effect of not applying cream over the abdomen was found to be significant in both the healthy group and the diabetic group (b = .64, t (110) = 5.57, $p < .001$; b = 1.52, t (25) = 2.78, p = .01). In the healthy sample, this was the only parameter that had an effect, however, in the diabetic group, the foetal abdominal circumference was found to have a significant interaction (b = 0.023, t (21) = 2.23, p = .04). A significant difference in the temperature means between the two clinical groups was found (F (1, 83) = 21.116, $p < 0.001$, η^2 = 0.203). A prediction model was based on the significance of the variation of the pool depth being influenced by the difference between the wall temperature and the quadrant maximum temperature measured at 30 minutes, in the healthy patients, and 45 minutes in the diabetic group. Finally, the intra-rater reliability test showed excellent level of agreement between operators, for minimum, maximum and average readings between regions of interest on thermogram (ICC – 0.875, 0.831 and 0.868 respectively), whilst the test-retest reliability showed an excellent level of agreement for the maximum and average temperature readings within the ROI (ICC – 0.989 and 0.997 respectively).

4 Discussion

In this research two phases of trials were conducted to explore the relationship between abdominal temperatures in pregnant women measured using thermography and liquor pool depth measured by ultrasound. The experiments were conducted in a hospital, climate-controlled environment using an infra-red thermographic camera and software to record and process the thermal images and ultrasound measurements using standard clinical equipment. Each trial allowed for a 60 minute acclimatisation period over which data was acquired. In the main phase (Phase 2) patients were healthy subjects while the next phase (Phase 3) was conducted on participants diagnosed as being diabetic.

In general the results for both phases indicated a significant, positive correlation between abdominal temperature and pool depth and that acclimatisation profiles suggest optimal time points for thermal measurements. Although Phase 3 results were in line with those in Phase 2, some differences in outcome for the diabetic cohort were noted.

4.1 General Findings

One finding in the Phase 2 and 3 trials was that the mean temperature recorded on thermography was significantly different between temperature levels at every time point. This means that across ROI's, there was a significant difference between the minimum, average and the maximum temperature levels determined by the thermography software at the five time points used. Additionally, as mentioned in section 2.2.1.3.1, positive concurrent validity results between oral and flexion point temperatures were obtained. Both these results validate the thermographic software representation of temperature, thus making it an important baseline finding with regards to the use of thermography in the setting of the pregnant abdomen, using the proposed methodology described in section 2. Nonetheless this software representation of temperature has some limitations which will be discussed later.

A second finding was that there was no significant difference in mean temperature recorded at the four ROI's across sample patients. This justified the use of quadrants as

independent entities for the purpose of data collection and sample inference since on average, the quadrant location does not influence the temperature reading. This finding is consistent with both Simoes (2012) and Pereira (2016) whose studies described the thermal symmetry between the left and right sided ROI's in the abdominal region of pregnant women. While the finding shows that for this study all four quadrants are valid as independent data-points for thermographic measurement, it should however be noted that the overall the results also found no effect of a foetal lie interaction. This is consistent but suggests that future research on thermography and foetal position would probably require a different methodological approach. An example would be to record more lateral thermal views of the abdomen in order to assess foetal lie, which may result in more positive outcomes as shown in the study by Topalidou, Markarian, & Downe (2020).

In the following sections the results are discussed in detail in relation to each of the research questions posed by this study.

4.2 **Research Question 1**

This question asked if there is a significant relationship between the thermographic temperature level and the maximum depth of the liquor pool measured with ultrasound at the regions of interest (ROI's) in a pregnant patient and if there is, can this be quantified.

In Phase 2, this question was answered by the salient findings that significant correlations between the maximum temperature (Tmax) at 15 minutes and (Tmax) at 30 minutes and the pool depth were found. However these relationships both showed a weak positive correlation. Association was shown to be much stronger when the ambient temperature (temperature of the wall) was controlled for. This is in line with the formula for radiative thermal emission

$$E_{measured} \cong h_r \epsilon_{obj} (T_{obj} - T_{env})$$

adopted by Arens & Zhang (2006) and mentioned in section 1.1.5.5.4. When the difference between the quadrant temperature and the wall temperature (QWT) was correlated with the

pool depth, the strongest correlations where obtained with the maximum temperature level QWTtMax, at 15 minutes and at 30 minutes thus reinforcing the results obtained with raw absolute temperature values. This result clearly indicates a relationship between the depth of the liquor pool measured on ultrasound and the temperature at that same ROI as measured on thermography. Furthermore, the results suggest which temperature level readings (QWTtmax) and which time values (15 minutes and 30 minutes), provide the strongest associations. The linear regression predictive model further showed that for the study sample, QWTt30max is a statistically significant predictor of pool depth with the higher the temperature difference, the larger the predicted pool depth.

As a tentative explanation for this association between the liquor pool depth and the abdominal temperature measurement, a proposed mechanism is being suggested. This is based on a study by Schroder & Power (1997). In their paper, they adopt a heat-engine approach to the thermodynamics of the foetal-placenta system, describing the foetal metabolism as the 'engine' and the placenta and liquor as the 'radiator'. The heat produced by the foetus is dissipated mainly via the placenta and then to a lesser degree, across other foetal membranes including the amniotic fluid, as was discussed in section 1.1.2. The principle finding of their study was that the foetal thermoregulatory mechanisms were strongly tied to the maternal temperature variations and that neonatal mechanisms for thermoregulation are effectively inhibited in utero. In their study, they also stated that the specific heat capacity of human blood was 85% that of water (3.6 J ml^{-1} $^{\circ}C^{-1}$ vs 4.2 J ml^{-1} $^{\circ}C^{-1}$ respectively). In the foeto-placental unit this implies that deeper liquor pools have a higher specific heat capacity than the locations with more foetal or placental tissue. This observation may support the hypothesis that the underlying pool depth determines the skin temperature change profile during and after a period of acclimatisation due to the different specific heat capacities of the constituents of the gravid uterus per unit volume underlying the abdominal wall.

Figure 51 illustrates this concept. Supposing the basic components in the pregnant uterus are the placenta, the foetus and the liquor, and assuming for the ease of calculation, that the total volume in each quadrant is 1000mls and taking the specific heat capacities as 4.2 J ml^{-1} $°C^{-1}$ for the liquor and 3.6 J ml^{-1} $°C^{-1}$ for the placenta and foetus, the diagram shows the resulting specific heat capacities for hypothetical volumes of the components for each sector. It can be observed that in this example the RLQ has the highest total specific heat capacity as it is within this quadrant that there is the largest assumed volume of amniotic fluid and hence the deepest liquor pool. This reasoning should also be valid for each ROI independently. Just as the results (section 3.3.2) showed a significant correlation between pool depth and temperature over all locations, grouping correlations by ROI should show

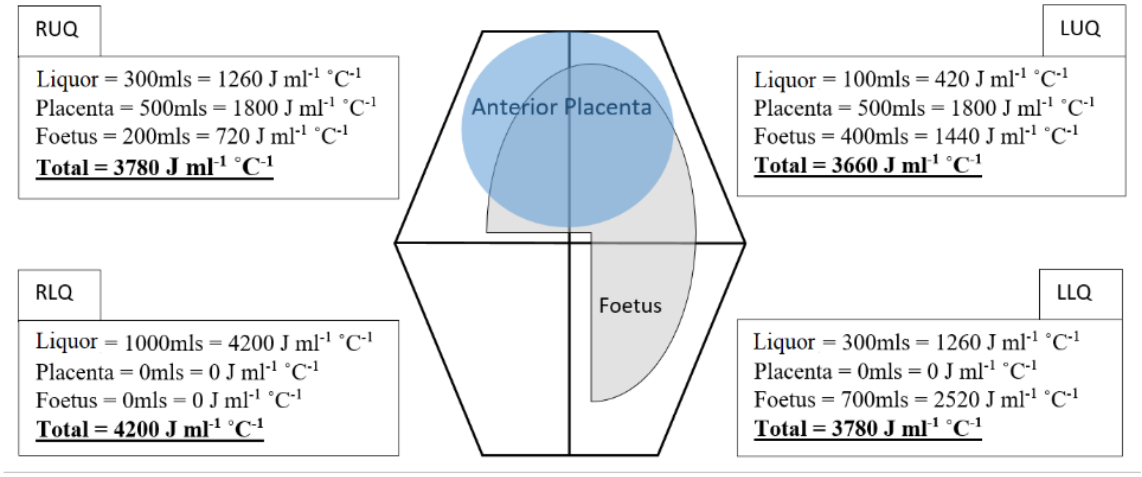


Figure 51. Specific gravity per sector in uterus

similar correlations for each group analysed separately and indeed does so, as shown in Table 16 (LLQ excepted because missing data rendered the sample insufficient), in support of this reasoning. Now consider Figure 52 which is a very simplified schematic cross section of two blocks showing heat flows from the foetus via the amniotic fluid, uterus, muscle and skin to the ambient air. The temperature gradient across the amniotic fluid ($T_f - T_a$) is the same in each block at a relatively constant 0.25 – 0.3°C (H. P. Laburn et al., 1992). This temperature

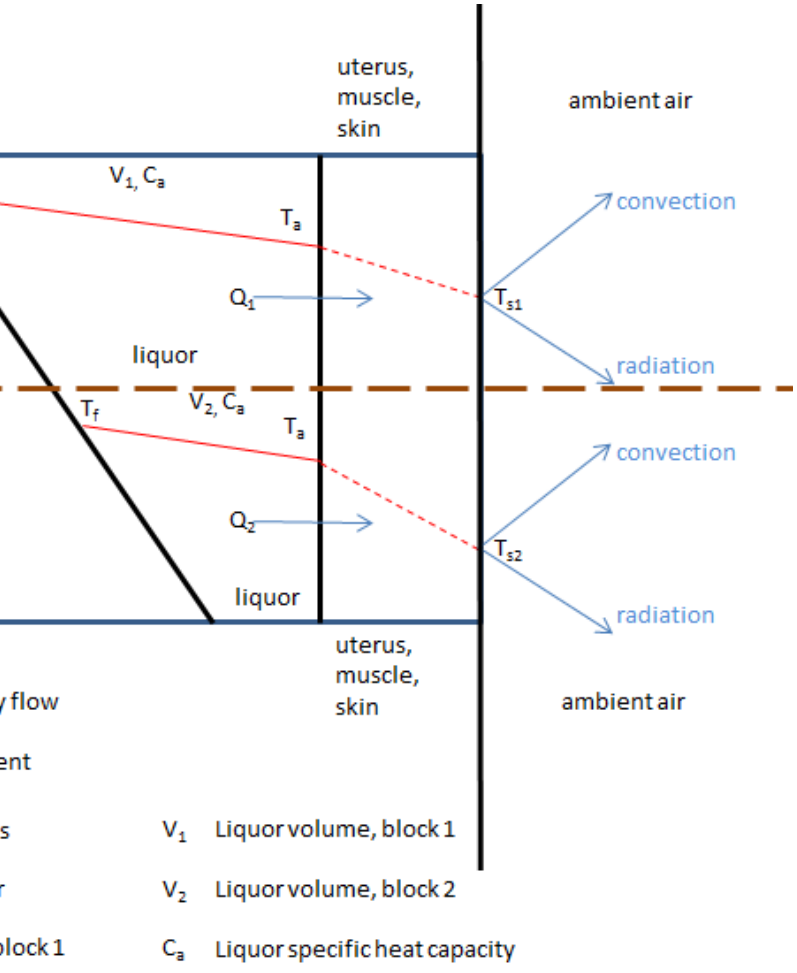
correlation between Pool Depth and QuadWallTt30max			
Location	***r***	***ρ***	***N***
RLQ	.491	.02	22
RUQ	.593	.002	25
LUQ	.706	.000	22

Table 16. Temperature to pool depth correlation per quadrant

difference results in heat energy flowing from the fluid to the maternal tissue, however the energy, which is given by

$$Q \propto VC(T_f - T_a)$$

is larger in block 1 because $V_1 > V_2$ and hence $Q_1 > Q_2$. The inference is that, since block 1 has a larger amount of energy flowing into the maternal tissue, disregarding all other heat sources, the skin temperature sustained in this block during acclimatisation and in equilibrium will be higher than the skin temperature for block 2 so $T_{s1} > T_{s2}$. Again the block having the deepest liquor pool will



schematic

show the highest temperature on thermographic temperature measurement when compared to the other blocks.

It has to be said that this model is an over-simplification for the sake of illustration because there are other sources of heat in the maternal tissue and the anatomical geometry leads to very complex heat flows. Nonetheless, the model serves to demonstrate how the specific heat of the liquor and the pool depth contribute to the marginal level of skin temperature. As further support for this hypothesis, a mixed (1x5x1) ANOVA was carried out for the 'average' temperature level at the 5 time points with 2 levels of pool depth as a between subjects factor. Pool depths were classified into two groups, pool depth > 4.7, N = 33 and pool depth =< 4.7, N = 39. The analysis showed that there was a significant difference between average temperatures for these two groups, $F(1, 70) = 5.05$, $p = 0.028$, $\eta^2 = 0.067$ with the mean temp for pool depth > 4.7 ($M_{\text{pool depth > 4.7}} = 31.15$) being significantly higher than the mean for the pool depth =< 4.7 group ($M_{\text{pool depth =< 4.7}} = 30.69$). The acclimatization profile for the two groups obtained in the supporting analysis is shown in Figure 53.

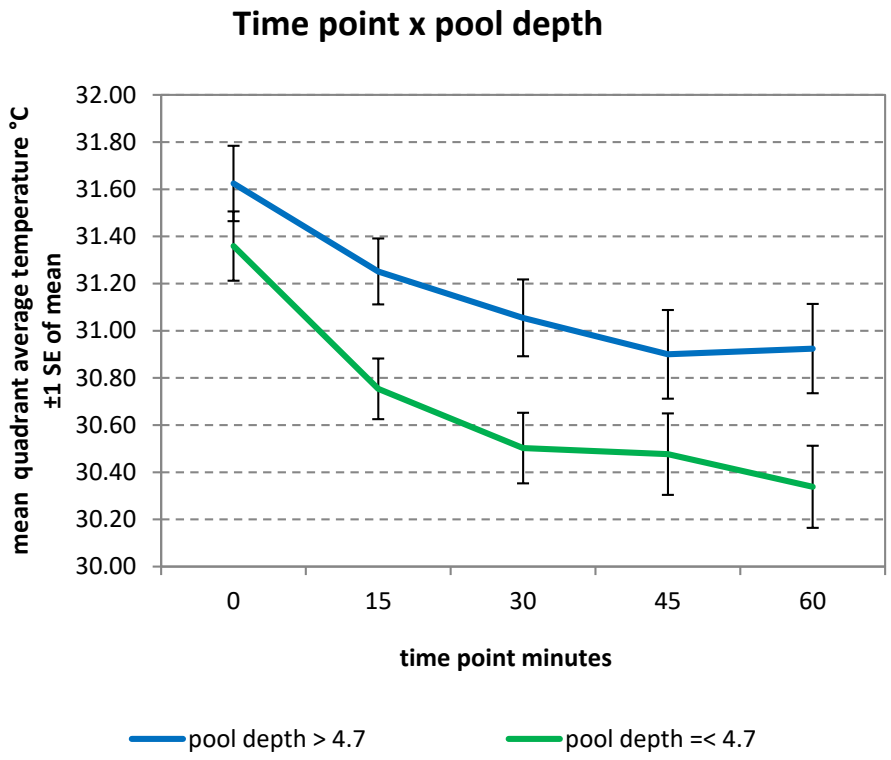


Figure 53. Mean temperature per time point for 2 level pool depths

Results for the diabetic cohort in Phase 3 showed general alignment with those of Phase 2 – correlation between temperature and pool depth was also indicated as seen in section 3.6.2. However here, the smaller sample size rendered the data noisier and results are only indicative. Nonetheless, although the incidence of correlation at time points and temperature levels was more limited, it can be reasonably suggested that the research question for the diabetic cohort was also answered positively. The impact of the diabetic condition on the results is further discussed in section 4.5 - Research Question 4.

For both phases the relationship between temperature and pool depth was quantified by the *r* values resulting from the correlation analyses but was also modelled in the regression equations for each cohort. The slopes of the regression lines are different, with the diabetic cohort having a steeper line plausibly because the temperature values for the two conditions are taken at different time-points and temperature levels – Phase 2 used QWTt30max while Phase 3 used QWTt45avg. This was because while for Phase 2 the most effective factor was identified via the stepwise backward regression analysis, the small sample size in Phase 3 meant that choice for the predictor factor was very limited and was judged by inspection. Nonetheless, allowing for these differences, the regression lines deliver similar pool depth predictions within the CI's of each line. This agrees with the finding that the clinical condition has a moderating significant interaction effect on QWTt45avg in predicted pool depth showing the diabetic condition having a higher influence albeit with a wider 95%CI.

It is also noted that the linear regression prediction intervals (PI's) for both cohorts at the mean temperatures are ±1.32 and ±1.74 cm for Phase 2 and Phase 3 respectively. This suggests that although the linear regression yields significantly predictive relationships for these samples, additional work needs to be done to reduce the PI's in order for the thermographic method to achieve expected clinical precision.

4.3 Research Question 2

This question asked if an acclimatisation period prior to thermographic measurements improved relevance of readings and if so what is this optimum acclimatisation period.

This equilibration period is an important aspect of any protocol in thermographic studies of humans that if standardised would also allow results from different studies to be properly compared. In various studies the chosen acclimatisation time frame seems to be arbitrarily chosen or based on what was used in previous work. Throughout the literature there are various proposed time periods ranging from as little as 5 minutes to 30 minutes (Fernández-Cuevas et al., 2015; Lahiri et al., 2012). In the few existing studies of thermography carried out in pregnant women, the acclimatisation period used was that of 15 minutes (T. Pereira et al., 2016; Simoes et al., 2012). Consequently, the study research question assessed the necessity of acclimatisation and the length of an adequate acclimatisation period when carrying out thermography over the pregnant abdomen.

In both phases of the study the patients' abdomen was exposed to the ambient room temperature of around 22°C. Findings from Phase 2 show that the mean abdomen temperature recorded on thermography was significantly different between time points up to the 30 minute reading. There was no significant difference between the temperatures recorded at 45 and at 60 minutes. This result is somewhat as expected as cooling follows Newton's Law that heat lost is proportional to the difference between ambient and body temperature (ASHREA, 2013). Specifically for this study environmental conditions the result indicates the effect and extent of acclimatisation, in that there is a sharp drop in mean temperature during the first 15 minutes as the outer skin cools relative to the ambient temperature and thereafter the temperature decline is less pronounced and indeed not significant after 45 minutes.

The research also analysed temperature decline of the different temperature levels recorded (minimum, maximum and average) across time points where they were found to

differ significantly. It was the maximum temperature measurements that sustained the least change over the first 15 minutes and least overall decline of temperature recording on thermography. Indeed the statistical analysis determined that the maximum temperature level at close to the 30 minute time point (QWTt30max) was the factor that contributed the highest R^2 in linear regression to predict pool depth.

These outcomes indicate that acclimatisation does matter in human thermographic studies and that there is an optimal point when temperature readings should be taken. They suggest that temperature decay in the cohort occurs most steeply during the first 15 minutes, reaching the most stable temperature at around 30 minutes. It also shows that taking the maximum temperature readings on the thermographic software will provide the least variability in temperature decline. The results suggest that for similar environmental conditions as this study (especially the constant ambient temperature level), an acclimatisation period of 30 minutes and taking the maximum temperature readings at the ROI would be a reasonable proposal for standard practice. These findings and elaborations in future work can be an important input for the development of protocols when using thermographic imaging in a healthy pregnant population.

Analysis of the acclimatisation period was also carried out for the diabetic group. There was a significant difference between time points up to the 45 minute reading. There was however, no statistically significant difference between the 45 and 60 minute temperature readings. This was in line with the correlation found using the Pearson analysis, where the most significant relationship between liquor pool depth measurement and the temperature difference between the wall temperature and the point temperature was found at 45 minutes. However, due to the small sample size these results should be interpreted with caution. This time point therefore can only be taken as a possible indication of an acclimatisation period within this pregnant diabetic sample. That being said, a longer equilibration period would be in line with the literature regarding the physiological

thermoregulatory changes within this group of patients. Other authors have suggested that there is a progressive deterioration in the sensory nerve pathways in diabetic patients, albeit the age of the patients has to be given due consideration. This results in affected thermoreceptors and mechanoreceptors, and causes an impaired skin blood flow and vasodilatory responses as part of their thermoregulatory haemostatic processes (Bharara et al., 2006; Kenny et al., 2016). Consequently, although the results need to be seen as an indication of the acclimatisation period required in this group, it is an interesting finding nonetheless in that it shows a potential effect of the diabetic state of the patients on this equilibration period.

4.4 Research Question 3

Here the influence of maternal and biological parameters on correlations of temperature and pool depth was explored and asking how these intrinsic and extrinsic factors affect this relationship.

A moderation analysis was carried out to assess if any bio-medical parameters recorded that relate to the mother and to the pregnancy had any effect on the relationship of the thermographic temperature and the liquor pool measurements. A statistically significant interaction of temperature and moderator in predicting pool depth would indicate a significant moderator influence.

The results for the Phase 2 trials confirmed that the application of cream to the abdomen was the only factor that had a significant interaction with temperature in predicting the pool depth. Specifically, it was found that there was a significant interaction of a dichotomous factor ‘cream’ with the temperature predictor. This finding was also confirmed in the Phase 3 results, where there was a similar significant interaction. The results showed that with cream applied, temperature could no longer be used as a significant predictor of pool depth. This is in line with previous studies. Bernard, Staffa, Mornstein, & Bourek (2013) showed that application of hydration cream results in statistical significant differences in the

temperature readings on the surface of treated and non-treated hands even up to 10 minutes after application. Their proposed explanation for this effect is that the cream lowers the skin emissivity thus causing the skin to radiate less energy, resulting in the temperature recorded by the camera to be reduced. Indeed, the results of the current study show that when cream was applied, thermographic readings are no longer significant predictors of pool depth. This suggests that application of cream alters the radiation of energy on the skin surface leading to misreading of temperature and thus making the use of cream undesirable for thermographic measurements.

In both Phase 2 and Phase 3 results, there was a non-significant effect of maternal skin type, which is in line with previous studies where no difference in emissivity was found between different skin colour types (Steketee, 1973). There was also no significant effect with regards to placental location or foetal lie, which is contrary to the expectations proposed in older research where placental location was assessed, not very successfully at the time, by using thermography (Liu & Blackwell, 1979; Millar, 1966). The advancement of both ultrasound and thermographic technology could be a reason for the differing results. Additionally recent research (Topalidou et al., 2020) has observed that lateral views yield improved assessment of foetal and placental location, so the frontal viewpoint adopted in the current study could have contributed to reducing possible effects of these factors.

An interesting result is that BMI and the mean abdominal thickness were not found to be influencing factors in both the healthy and the diabetic cohort. This could be due to the fact that BMI more than 40 kg/m^2 were excluded, and there were no extremes of abdominal thickness and BMI in the data collected. These findings also do not support the results in the study by Pereira et al. (2016) where there was a significant negative correlation between the average body temperature recorded and the BMI, meaning that as the BMI increases, the temperature recorded is less. In that study average body temperature encompassed all areas of the body including for example the breast where increased vascularisation during pregnancy

would result in an increase in the total body temperature profile. In the current study however, it is the abdominal temperatures alone that are being measured and analysed with the BMI, and this could be the reason for the difference in results. Indeed, a recent study (Topalidou et al., 2020) that focused only on thermography of the abdominal area in pregnant women also found no significant correlations of temperature with these two factors.

Pereira et al. (2016) and Simoes et al. (2012), both found a correlation of temperature with maternal age. In this study however, this parameter was not found to have a significant effect in both Phase 2 and Phase 3. The reason for this may be because of the lack of variation in ages within the population that was studied, as well as once again the distinction between the body area where the mean temperature readings were taken (total body in the comparative study vs the abdominal region in the present study).

With regards to abdominal striations, systolic blood pressure, diastolic blood pressure and pulse, there was no significant interaction found in both the Phase 2 and Phase 3 studies.

Of interest is the result in the Phase 3 study with regards to the abdominal circumference (AC) measurements, where these were found to have a significant interaction with QWTt45avg on the pool depth measurement. It was found that for this sample, at an AC of 29 cm the relationship between temperature and pool depth becomes significant and increases in significance and strength as AC increases. To this researcher's knowledge this possible effect has not been addressed in other studies in thermography of pregnant women. This parameter was not found to have a significant interaction in the healthy cohort of patients in the Phase 2 study. The use of the AC measurement on ultrasound as a predictor of macrosomia in diabetic patients has been investigated and confirmed extensively (De Reu, Smits, Oosterbaan, & Nijhuis, 2008; Rosati, Arduini, Giri, & Guariglia, 2010). There have also been studies which showed that the insulin levels within the amniotic fluid could be correlated to the foetal AC measured on ultrasound, emphasising the importance of this sonographic measurement (Schaefer-Graf et al., 2003). A more recent study carried out by

Sovio U, Murphy HR, & Smith GC. (2016) was carried out as part of the Pregnancy Outcome Prediction study at Addenbrooks Hospital, Cambridge. They found that patients who had an AC >95th centile and an HC:AC ratio of <10th centile at 28 weeks had double the risk of developing GDM in the pregnancy. This was not found to be a significant finding at earlier gestation scans, and resulted in the proposal of OGTT tests to be undertaken at 24 weeks rather than 28 weeks as is currently done on the NHS. This indicates that not only do AC measurements predict hyperglycaemic maternal states during pregnancy, but it does so as early as 28 weeks' gestation. In the current study, the gestational ages studied started from 28 weeks in both groups. The findings of the research previously discussed, especially the latter, could explain why the larger abdominal circumference measurements in the diabetic patients (M_{AC} = 28.9 cm) had an effect on the temperature relationship with the pool depth, whilst this was not the case in the healthy sample (M_{AC} = 23.5 cm).

Although further research is required in this area with a larger and more varied cohort, potentially however, if standards for temperature readings by thermography in pregnant patients are created in relation to gestational age, the finding of mean abdominal temperature lower than this standard could be an indicative triage mechanism for patients predisposed to diabetes in pregnancy.

Within the diabetic cohort, the use of insulin and of OHA's was also analysed, however no interaction was found with both treatments and QWT45avg on pool depth. The small sample size and thus even smaller numbers of patients on the different treatments could have resulted in too low a statistical power to uncover such effects.

In general, although this study did find specific moderation effects of maternal and biological parameters on correlations of temperature and pool depth it is clear that further research in this regard is required with bigger samples incorporating more variation of pregnant participants.

4.5 Research Question 4

This question asks if the same relationship between the thermographic temperature level and the maximum depth of the liquor pool measured with ultrasound at the ROI's exist in diabetic as with normal patients. The pertinent findings for the diabetic cohort were discussed in the previous sections together with Phase 2 result considerations. However the results comparing differences between the diabetic and the healthy group of patients merit further discussion.

The statistical comparison of the mean temperature over the different time points and levels per clinical condition showed a significant difference of approximately 1˚C, the diabetic group having a lower mean temperature than the non-diabetic. There was also a significant difference between temperature levels per condition interaction, with the diabetic group having a consistent lower mean temperature than the non-diabetic. These findings are in agreement with those of Kenny, Sigal, & Mcginn (2016) that discussed the effect of diabetes on thermoregulatory mechanisms. They describe how in Type 1 diabetic patients, the hyperinsulinaemic state causes a greater skin blood flow which enhances heat loss. This is combined with hypo-glycaemia mediated reduction in shivering thermogenesis. Reference is also made to Type 2 diabetic patients with reduced cutaneous vasoconstrictor response to cold stress. This research suggests that a difference in skin temperature profile between the diabetic and the healthy cohort would be expected in the current study. It would be anticipated that in diabetic patients, having an altered thermoregulatory function would result in them dissipating more heat from the skin, because of a delay in vasoconstriction of peripheral blood vessels when exposed to the surrounding environment, thus having lower mean temperature readings than the non-diabetic group.

Furthermore, as discussed in section 4.4 above a higher mean AC would be expected in diabetic patients. Applying the notion proposed earlier in section 4.2, if AC increases then V would decrease and disregarding all other heat sources, would result in a lower skin

temperature T_s during and after acclimatisation. This was indeed the finding in the current study.

4.6 Strengths of the Research

The methodology used in Phase 2 and Phase 3 was refined after the broader pilot study was carried out. The various changes made all aimed to strengthen the research in order to make it more reproducible, meaning that the methodology was devised in such a way so as to enable future research in this field to be standardised in the way it is carried out. To date, the sample size that was recruited for the study was the largest when compared to previous studies, at more than 40 patients for the healthy cohort (T. Pereira et al., 2016; Simoes et al., 2012; Topalidou et al., 2020). A convenience sample was obtained in the recruitment phase meaning that the collection was inexpensive and quicker than other forms of sampling. The strict inclusion and exclusion criteria allowed for the collected sample to be more representative of the larger healthy pregnant population.

One of the main strengths of this research is the fact that thermography was shown to be a technique with high reliability, with a very short learning period for the operator. This effectively means that it is more cost effective to train operators for this technique when compared to ultrasound. This means that, as is discussed later in the chapter, this modality can be set up in areas where ultrasound is not so readily available with short duration training at cheaper costs.

Another strength is the inclusion of an acclimatisation study. The analysis of the data showed with confidence the proposed acclimatisation time in a healthy group of pregnant patients as well as an indication of the time required in diabetic groups, as discussed in section 4.3 above.

This study is the first documented thermographic study which specifically addressed a diabetic pregnant cohort. The results allowed for the comparison of a healthy sample of pregnant participants with the diabetic group and found significant temperature differences

between the two groups. This is an important finding which can be used as a basis for further research with clinical application as is discussed further in section 4.8.1.4 below.

4.7 Limitations of the Research

Although from the main results of this study it can be inferred there is clear evidence of a systematic effect between temperature and pool depth in both Phase 1 and Phase 2 trials, however the magnitudes and variances of standard error values also suggest some potential limitations in the adopted methodology.

4.7.1 Methodological Limitations

Outcomes from a pilot study served to refine the subsequent research design wherever and to the extent possible. For example a standardised process and environment were adopted and kept similar for both Phase 2 and Phase 3 to reduce inconstancies between the two studies. Nonetheless, practical constraints introduced some limitations to the study design and these are discussed in the following sub-sections.

4.7.1.1 Recruitment

Recruitment for this study was a definite limitation. This was to be expected as patients are increasingly more sceptical with various procedures involving new technologies during pregnancy. In fact for Phase 2 only 40% of patients contacted showed an interest in participating and although these were reassured by recruiters about the technology being investigated the overall recruitment yield was only 15%. Recruiting the diabetic cohort is intrinsically difficult due to the low incidence of diabetes in the pregnant population as already discussed in the section 2.7.4.2 above. In order to improve the rate of recruitment here, there was no distinction between the ‘type’ of diabetic patient recruited – gestational diabetes, type 1 and type 2 diabetics. Although this was acceptable in an exploratory study such as the one carried out, it is a factor that should be addressed in future work in order to look at any differences in the thermographic measurements within these three sub groups.

Due to these recruitment issues, the trials were carried out over an extended time span of over 12 months which potentially lead to other logistic limitations. Grouping participants in consecutive sessions was rarely possible because of patient/researcher availability and thermography equipment availability as this was loaned from the Centre for Biomedical Cybernetics at the University of Malta on a per-occasion basis. Since this meant that the study environment had to be setup numerous times and although all effort was made to replicate the setup each time, unavoidable small changes could creep in and be reflected in random errors in the resulting data.

For future work given similar circumstances, less 'noisy' results could probably be attained by going for a smaller sample of patients that can be trailed over a short intense period of sessions and acquiring and processing a higher number of data points per session by utilising dynamic rather than static imaging as described in section 1.1.5.7. The frequent rotation of patients will help achieve similarity and minimize random differences in their processing.

4.7.1.2 Equipment

Infrared thermography as used in this study requires small temperature changes to be detected under unfavourable conditions of spurious thermal radiation in the environment and vascular tissue near the skin surface. The cameras used in the current study were medium-cost mobile thermal cameras with acceptable image quality but having the lowest detector resolution recommended (320x240) for adequate human thermal data medical work (Quesada et al., 2017). Higher detector and thermal resolution cameras would afford more accurate calculations of temperatures and would be recommended for future research.

4.7.1.3 Study environment

As mentioned in the Methodology section above, although IR thermography imaging has high dependence on the environment (Usamentiaga et al., 2014) there is no one agreed

standardised value for ambient temperature and relative humidity of the study environment (Fernández-Cuevas et al., 2015). In this study the climate controlled environment in the study room was retained at approximately 21°C which is in the preferred range of many other studies. The results indicated that controlling for the mean room (or wall) temperature by subtracting its value from direct abdominal thermographic readings resulted in strong statistically significant correlations with pool depth.

This presents a limitation of the study in that it cannot be compared to a baseline wall temperature setting and it cannot as such establish if a higher or lower room temperature would have decreased standard errors in temperature data and thus improved the results. Future work could address this by investigating the effects of various abdomen-room temperature differences on thermography obstetric applications.

4.7.1.4 Patient Positioning

The patient was required to lie in a supine position at approximately 45 degrees for 60 minutes duration. As discussed in section 2, during pregnancy one needs to be aware of the risk of caval compression which can lead to vasovagal responses. In fact during the study one patient's data was excluded from Phase 2 as she did suffer a vasovagal early on in the session (the patient was tended to by the researcher who is a senior obstetrician and after a few minutes she was recovered and sitting upright). To de-risk the protocol, the pregnant participants were not strictly required to sit at a 45° angle – it was only suggested and could be adapted to the patient's comfort. To mitigate this limitation, the only requirement was that the entire abdomen was visible on the thermographic imaging. Still, slight orientation differences between patients could potentially have increased random errors in the data. In future work it is suggested that lateral ROI's in addition to the 4 planar ROI's used in this study are also included. Indeed, other researchers have suggested a 5 viewpoint synchronised IR thermography video recording as a standard (Topalidou et al., 2020).

4.7.1.5 ROI selection and thermal data analysis

In this study, the thermal data was processed by manually selecting the ROI using the IR camera software for each thermographic image. The area of the ROI was based on the assumption that the uterus and its contents lie within the more central area of the abdomen and so the researcher selected the four ROI's most medial to and around the umbilicus. The software is restricted to selecting circular or oval ROI's which limited the researcher in the area that can be selected. For this reason standardised ROI shapes as used in other studies such as by Pereira et al. (2016) and Simoes et al.(2012) who selected triangles over the LUQ and the RUQ and ellipses over the LLQ and the RLQ were not possible. The dimensions of the ROI have an implicit effect on the thermographic measurement since hot-spots have a bigger effect on small sized ROI's that will show an increased temperature (Quesada et al., 2017). This is an inevitable limitation in this study which could have slightly affected the temperature measurements processed. To an extent this limitation was controlled for by carrying out a reliability study of the use of the thermographic software to select ROI's which showed that intra-rater and inter-rater tests had high ICC values, implying that the selection of the ROI's was consistent both between operators and with one operator on different days.

ROI selection is a critical topic in obstetric use of thermography and this limitation will be addressed in the future work discussion later in this chapter.

As in most reported research using thermography in obstetrics, this study was limited to using static imaging of ROI's. This is a limitation because static images only acquire spatial data whilst the mother-foetus system under study is a dynamic one which changes in both time and space. Furthermore foetal movements may manifest as changes in the patterns of abdominal surface heat maps (Topalidou et al., 2020). Using dynamic thermography with appropriate thermal image processing software in future work as described by Falzon et al., (2018) could mitigate this limitation.

4.8 Future work

The study results and limitations give direction to a number of possibilities for future development and research some of which were suggested in the section above. Generally, it is proposed that a protocol based on the current research, using a 30 minute acclimatisation time should be followed in future developments of this experiment. This will allow information analysed to be compared to the results of this study and to continue to build on the current work.

4.8.1.1 ROI selection

Currently, the selection of ROI's is done mostly manually, and the shape of the ROI is either drawn freehand on the image or provided by the thermography camera software which for non-specialised camera systems is mainly geared for industrial applications rather than human body measurements. Also the mean temperature readings are recorded as a mean of all the pixels within the ROI selected (Quesada et al., 2017). A future study could further this current study by looking into the development of ROI shapes which would take into account the curvature of the gravid abdomen, and integrate other methods of temperature calculation within the ROI. Two potential approaches can be suggested.

One recently proposed method is the 'Tmax' method which is a mathematical method that uses the same number of pixels for calculations within any operator selected ROI. The advantage is that it normalises different patient anatomical sizes (Ludwig et al., 2014).

The other approach mentioned in section 4.7.1.5 above is used with dynamic thermography and involves a registration technique of the thermography video frames of the ROI that aligns spatial points in the frame sequence. This method does not require the use of physical markers and thus reduces subjectivity (Ciantar et al., 2018).

4.8.1.2 Diabetic Patients

Another line of investigation for research could be the expansion of the findings in Phase 3 of the current study. As was already stated, the results of this study phase should be interpreted with caution. However as preliminary findings, they do provide data that is worth expanding. Since recruitment of a larger sample size with a division of groups by treatment would be problematic locally for reasons already mentioned, it would be sensible to undertake such project in association with other medical/academic institutions in the EU.

4.8.1.3 Extremes of Pool Depth

In the current study, the extremes of pool depth were excluded when the data was screened, and they were seen as outliers as none of the patients suffered polyhydramnios or oligohydramnios. A future study could explore these extremes of pool depth and their relationship with thermographic temperature measurements. According to the RCOG Green top Guideline, oligohydramnios is defined as a single deepest liquor pool of ≤ 2cm or an AFI of ≤ 5cm. The incidence of an AFI of ≤ 5cm in a low risk population is 1.5% (Royal College of Obstetricians and Gynaecologists, 2013). Polyhydramnios occurs in 1 per 100 pregnancies according to the Foetal Medicine Foundation, and is defined as single deepest vertical liquor pool measurement of 8 to 16cm depending on severity reference. Undertaking such a study, would probably face difficulties in recruitment in view of the low prevalence of both conditions, however the results would be relevant especially if translated into a future application of early detection of such conditions.

4.8.1.4 Clinical Application

The findings of the present research suggest a future role for thermography as an adjunct device in obstetrics to investigate the pathology of pregnancy. The empirical findings of the research provide a new understanding of how this method measures the temperatures over the abdominal skin of the gravid uterus, and how the underlying pregnancy unit

correlate to these temperature patterns. This provides confidence in future work which can ultimately result in a non-invasive device that can be utilised to triage patients for various clinical conditions. With more research dedicated to specific conditions in obstetrics, such diabetes, hypertension and placental dysfunction, standardised protocols and temperature measurement databases corrected for these groups can be developed. This together with the integration of the thermal map and the ultrasound image, as well as more targeted thermographic software for use in obstetrics, could result in a new method of predicting conditions in pregnancy, with a reduction in operator bias. The early detection of such conditions would lead to earlier treatment and in turn a reduction in maternal and foetal morbidity and mortality.

Specifically, the findings pertaining to the diabetic cohort suggest that following further research, thermography could be used as a tool for early detection of diabetic tendencies in pregnancy. As seen in section 4.4, the foetal abdominal circumference is a known predictor of development of gestational diabetes. As proposed in this same section, recording a lower temperature than set standardised temperatures for that same gestation and clinical group could be a novel way to predict the predisposition for diabetes, preceding the ultrasound findings of larger abdominal circumference. This could be of value in regions where ultrasound is not readily available and thermography being an overall cheaper modality would be a viable option in such situations.

4.9 Conclusion

The current study showed clear evidence of correlation between thermographic temperature measurements and liquor pool depth measured on ultrasound for both non-diabetic and diabetic pregnant women. Regression lines were computed for temperature as a significant predictor of pool depth. It was shown that the patient's clinical condition moderated the effect of temperature as a predictor of pool depth, that application of cream in both study groups has an influence on this relationship but foetal abdominal circumference

had a significant moderation effect only in the diabetic group. The mean temperature over time points and levels per clinical condition showed a significant difference of approximately 1˚C, the diabetic group having a lower mean temperature than the non-diabetic. An acclimatisation period of 30 minutes for the healthy cohort and 45 minutes for the diabetic cohort was suggested as a result of the study findings. The limitations of the study were identified and discussed while suggestions for extending current work in future directions were recommended.

Carrying out research in this field is important in order to explore the possible role that thermography could have in the area of obstetrics as it is a promising technique mainly due to its non-invasive and non-contact properties. The novelty of this research and its results show that although further studies are required to determine optimal methodology for thermography research on pregnant women, it can be an additional research tool in the area of obstetric health studies. An added facet of this study was the insight that it provided on the thermographic data outcomes in the clinical group of diabetic pregnant patients.

From a practical standpoint, this study has added to the understanding of how in a stable environment and following a standardised protocol, infra-red thermography shows promise to be a novel device in the field of obstetrics and for different clinical groups, which will provide added information with regards to the pregnancy.

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Appendix A – Data fields

Phase 1 (Pilot phase) compiled data

i. Depth of each liquor pool for each subject
ii. Depth of the deepest liquor pool for each subject
iii. Amniotic Fluid Index for each subject
iv. Temperature of the coldest thermographic area for each subject
v. Temperature at umbilicus for each subject
vi. Skin thickness as measured on ultrasound at 12, 3, 6 and 9 o'clock for each subject
vii. Average skin thickness for each subject
viii. Skin thickness of umbilicus as measured on ultrasound for each subject
ix. Striae presence
x. Presentation (Lie)
xi. Gestation
xii. Placental site

Phase 2 and Phase 3 compiled data

Ultrasound data

i. Right lower quadrant liquor pool depth
ii. Right upper quadrant liquor pool depth
iii. Left upper quadrant liquor pool depth
iv. Left lower quadrant liquor pool depth

Thermographic data

i. Temperature at Umbilicus at 0 minutes
ii. Temperature at Quadrant at 0 minutes (minimum)
iii. Temperature at Quadrant at 0 minutes (maximum)
iv. Temperature at Quadrant at 0 minutes (average)

v. Temperature at Umbilicus at 15 minutes

vi. Temperature at Quadrant at 15 minutes (minimum)

vii. Temperature at Quadrant at 15 minutes (maximum)

viii. Temperature at Quadrant at 15 minutes (average)

ix. Temperature at Umbilicus at 30 minutes

x. Temperature at Quadrant at 30 minutes (minimum)

xi. Temperature at Quadrant at 30 minutes (maximum)

xii. Temperature at Quadrant at 30 minutes (average)

xiii. Temperature at Umbilicus at 45 minutes

xiv. Temperature at Quadrant at 45 minutes (minimum)

xv. Temperature at Quadrant at 45 minutes (maximum)

xvi. Temperature at Quadrant at 45 minutes (average)

xvii. Temperature at Umbilicus at 60 minutes

xviii. Temperature at Quadrant at 60 minutes (minimum)

xix. Temperature at Quadrant at 60 minutes (maximum)

xx. Temperature at Quadrant at 60 minutes (average)

xxi. Temperature of the wall on thermogram at 0 minutes

xxii. Temperature of the wall on thermogram at 15 minutes

xxiii. Temperature of the wall on thermogram at 30 minutes

xxiv. Temperature of the wall on thermogram at 45 minutes

xxv. Temperature of the wall on thermogram at 60 minutes

Computed fields from thermographic data

i. Temperature difference between Temperature at Quadrant at 0 minutes (maximum) and Temperature of the wall on thermogram at 0 minutes

ii. Temperature difference between Temperature at Quadrant at 0 minutes (average) and Temperature of the wall on thermogram at 0 minutes

iii. Temperature difference between Temperature at Quadrant at 0 minutes (minimum) and Temperature of the wall on thermogram at 0 minutes

iv. Temperature difference between Temperature at Quadrant at 15 minutes (maximum) and Temperature of the wall on thermogram at 15 minutes

v. Temperature difference between Temperature at Quadrant at 15 minutes (average) and Temperature of the wall on thermogram at 15 minutes

vi. Temperature difference between Temperature at Quadrant at 15 minutes (minimum) and Temperature of the wall on thermogram at 15 minutes

vii. Temperature difference between Temperature at Quadrant at 30 minutes (maximum) and Temperature of the wall on thermogram at 30 minutes

viii. Temperature difference between Temperature at Quadrant at 30 minutes (average) and Temperature of the wall on thermogram at 30 minutes

ix. Temperature difference between Temperature at Quadrant at 30 minutes (minimum) and Temperature of the wall on thermogram at 30 minutes

x. Temperature difference between Temperature at Quadrant at 45 minutes (maximum) and Temperature of the wall on thermogram at 45 minutes

xi. Temperature difference between Temperature at Quadrant at 45 minutes (average) and Temperature of the wall on thermogram at 45 minutes

xii. Temperature difference between Temperature at Quadrant at 45 minutes (minimum) and Temperature of the wall on thermogram at 45 minutes

xiii. Temperature difference between Temperature at Quadrant at 60 minutes (maximum) and Temperature of the wall on thermogram at 60 minutes

xiv. Temperature difference between Temperature at Quadrant at 60 minutes (average) and Temperature of the wall on thermogram at 60 minutes

xv. Temperature difference between Temperature at Quadrant at 60 minutes (minimum) and Temperature of the wall on thermogram at 60 minutes

Biomedical and demographic data

i. Maternal Age (years)

ii. Gestational Age (weeks)

iii. Maternal Systolic Blood Pressure (mmHg)

iv. Maternal Diastolic Blood Pressure (mmHg)

v. Maternal Pulse (bpm)

vi. Maternal Oral Temperature (˚C)

vii. Maternal Body Mass Index

viii. Maternal Skin Type as described by the Fitzpatrick classification (Fitzpatrick, 1988) (presented as 1 = Type 1; 2 = Type 2; 3 = Type 3; 4 = Type 4; 5 = Type 5)

ix. Application or not of Cream (Phase 2: 1=Cream not applied, 2=Cream Applied; Phase 3: 0=Cream not applied, 1=Cream Applied)

x. Presence of Striations (1= Striations Present; 0= Striations Absent)

Computed fields from Ultrasound data

i. Placental Location (1 = Anterior placenta; 2 = Posterior placenta; 3 = Fundal placenta)

ii. Foetal Lie (1 = Cephalic presentation; 2 = Breech presentation; 3 = Transverse presentation)

iii. Foetal Abdominal Circumference (cm) (as measured on ultrasound using the Hadlock measurements)

iv. Maternal Mean abdominal wall thickness – the mean of all the measurements of the abdominal wall taken by ultrasound

Appendix B – Patient Communication

Recruitment communication (Phases 2 & 3)

Sinjura,

Jiena tabiba li naħdem fid- Dipartiment tal-Ostetrija u Ġinekoloġija u bħalissa qed nistudja għal PhD ma l-Universita ta Malta.

L-istudju tiegħi qed iħares lejn it-temperaturi taz-żaqq waqt li inti tkun tqila, speċifikament ta 28 il-ġimgħa tqala. Fil-proċess ta l-istudju, ser jittieħed stampa tat-temperaturi emessi minn żaqqek permezz ta kamera speċjali. M'hemm l-ebda xrays jew raġġi oħra ta ħsara involuti. Aħna sempliċiment ħa naraw is-sħana emessa miż-żaqq u wara nagħmlu 'ultrasound' tat-tarbija tiegħek. Dan kollu għandu jieħu xi siegha b'kollox.

L-Għan ta dan l-istudju huwa li nikkomparaw ir-'ritratt' tat-temperaturi fuq żaqqek ma dak li naraw fuq l-'ultrasound'. Aħna ħa nkunu qiegħdin inħarsu lejn l-ilma ta madwar it-tarbija, il-pożizzjoni tat-tarbija u anke tas-sekonda, u dak kollu li ħa jgħinha fil-proġett tagħna. Nisperaw li dan l-istudju iwassal għal metodu ġdid kif ninvestiġaw it-tqala biex inkunu nistgħu nindunaw b'ċerti problemi iktar kmieni milli diġa qiegħdin nagħmlu bħal issa.

Bħal kull studju ta dan it-tip, l-iktar ħaġa importanti huma intom il-pazjenti u l-għajnuna tagħkom. Għalhekk qed insaqqsikhom biex jekk intom interessati jew għandkom xi mistoqsijiet tistgħu tibgħatu email fuq martina.muscat@gmail.com jew cempluli fuq 99489999.

L-Għajnuna tagħkom hija importanti ħafna.

Grazzi!

Dr.Martina Schembri
Dipartiment tal-Ostetrija u Ġinekoloġija
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Prof.Y.Muscat Baron
Superviżur tar-Riċerka
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Dear mother-to-be,

I am a doctor working in the Department of Obstetrics and Gynaecology and am currently reading for a PhD with the University of Malta.

My study is looking at temperatures of the pregnant uterus at around 28weeks of pregnancy and involves taking a picture of the temperatures emitted by your abdomen with a special thermographic camera. There are no Xrays or other harmful rays involved. We simply pick up the heat emitted off your abdomen. After this we carry out an ultrasound of your baby. The process will take in total approximately one hour of your time.

The aim of this study is to compare the temperature 'picture' to the ultrasound image. We will be looking at the fluid around the baby, the position, the placental position and anything else that we can compare. We hope that in the end we will find a correlation between the two images and this will lead to a new way of assessing the fetus for routine checkups and to also pick up problems earlier than we already currently are.

Like all studies of this kind, the most important entity is the patient and their co-operation. For this reason, I urge you to please contact me if you are interested in taking part in this study. Your help is invaluable!

Please email me on martina.muscat@gmail.com, or contact me on 99489999 if you decide to partake in this study or if you have any questions about it.

Thank you for taking the time to read this.

Dr. Martina Schembri
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Appendix C – Consent

Consent form

CONSENT FORM (English)

Dear Madame,

The Department of Obstetrics and Gynaecology, the Department of Systems and Control Engineering and the Centre for Biomedical Cybernetics of the University of Malta would like to invite you to undertake a totally safe study during your pregnancy. The study is entitled Thermography in Pregnancy which means temperature studies in Pregnancy.

The commonest test your child will have throughout his life will involve checking his/her temperature. Until now this test could not be performed on the unborn baby due to several technical limitations. With significant advances in technology the Department of Systems and Control Engineering of the University of Malta is equipped with a temperature probe, which from a distance from your belly may measure temperatures related to your baby. The equipment is totally safe and functions by detecting the heat your body and your baby emits. The equipment is like a thermometer which can measure temperatures without touching your belly and in a passive manner - only collecting heat that is emitted from yourself.

All information collected during this study will be confidential, and will only be used for the purposes of this study. My supervisor and examiners may also require access to this data for the purpose of verification. All data will be utilised according to the GDPR Regulations (EU) 2016/679. Consent for this study is optional.

☐ I am willing to participate in the study

☐ I allow my data to be utilised for the purpose of the study and can be accessed by myself, my supervisor and examiners for the sole purpose of verification of the data if required

Signature ……..

(Name in block letters ……..……..)

I.D……..

Date……..

I …….. (Block Letters ……..……..) I.D……..have / have not obtained informed consent from ……..to have the Thermography test performed during her pregnancy. DATE……..

Formula ta' kunsens (Malti)

Sinjura,

Id-Dipartiment ta Ostetricja u Ginekologija, d-Dipartiment ta' 'Systems and Control Engineering' u c-Centru tal-'Biomedical Cybernetics' ta' l-Univesita' ta' Malta, jistiednuk biex taghmel studju li huwa kompletament "safe" waqt it-tqala tieghek. It-titlu tal-istudju jismu Termografija fit-Tqala li tfisser studju ta' temperatura waqt it-Tqala.

L-iktar haga medika li se tigi ccekjata waqt il-hajja tat-tifel/tilfa tieghek hija t-temperatura. S' issa t-temperatura tat-tarbija gol-guf ma setax tigi ccekjata minhabba prolemi teknici. Bl-avvanzi teknologici, issa d-Dipartiment tas-'Systems and Control Engineering' fl-Universita' huwa mghammar b'apparat li jista jiccekkja t-temperatura tat-tarbija tieghek minghajr ma tintmiss. Dan il-apparat huwa totalment "safe" u jiffunzjona billi bhal termomitru jiccekja s-shana hierga minn go fik u it-tarbija tieghek.

L-informazzjoni kollha miġbura matul dan l-istudju se tkun kunfidenzjali, u tintuża biss għall-finijiet ta 'dan l-istudju. Is-superviżur u l-eżaminaturi tiegħi jistgħu jeħtieġu wkoll aċċess għal din id-dejta għall-fini tal-verifika. Id-dejta kollha se tintuża skond ir-Regolamenti tal-GDPR (UE) 2016/679. Hija l-ghazla tieghek jekk tridx tippartecipa fl-istudju jew le.

☐ Jiena lest li nipparteċipa fl-istudju

☐ Jiena nippermetti li d-data tiegħi tiġi utilizzata għall-iskop ta 'l-istudju u tista' tkun aċċessata mir-ricerkatur, mis-superviżur u l-eżaminaturi involuti fir-ricerka għall-iskop uniku ta 'verifika tad-data jekk meħtieġ

Firma……..

Isem ……...……..

I.D……..

Data ……...……..

Jiena…….(block letters ……...…….)
I.D……..ircevejt / ma rcevejtx l-kunsens infurmat minn ……..biex isir it-test tat-termografija waqt it-tqala taghha.
Data……...……..

Appendix D – Questionnaire

Patient Questionnaire

Date: _____

Name: _____

Personal Code: _____

Ethnicity

Caucasian ☐ Asian ☐ African ☐

Outside temperature: _____ Core Temperature: _____ Room Temperature: _____

Have you used cream on your abdomen? YES ☐ NO ☐
If yes – when? _____

Have you gone sunbathing in the last 3 days? YES ☐ NO ☐
If yes – when? _____

Have you smoked any cigarettes today? YES ☐ NO ☐
If yes – how many? _____

1st Pregnancy? YES ☐ NO ☐

Previous Miscarriages? YES ☐ NO ☐

If yes: How many? _____

Medical Management _____

Surgical Management _____

Present Pregnancy

Last menstrual Period _____

Estimated Due Date by Ultrasound _____

Bleeding Early Pregnancy YES ☐ NO ☐

Admissions to Hospital YES ☐ NO ☐

Reason for admission

_____

_____

Reason for admission

_____

_____

Medications (Gestation started/ Gestation stopped)

Progesterone	YES ☐	NO ☐
Aspirin	YES ☐	NO ☐
Clexane	YES ☐	NO ☐

_____

_____

_____

_____

_____

_____

_____

Current Medical Conditions:

Hypertension	YES ☐	NO ☐
Hypothyroidism	YES ☐	NO ☐
Hyperthyroidism	YES ☐	NO ☐

Surgery during Pregnancy YES ☐ NO ☐

Past Medical History

Hypertension	YES ☐	NO ☐
Asthma	YES ☐	NO ☐
Hypothyroidism	YES ☐	NO ☐
Hyperthyroidism	YES ☐	NO ☐
Other	YES ☐	

_____

_____

_____

_____

Past Surgical History

Appendicectomy	YES ☐	NO ☐
Other	YES ☐	

_____

_____

_____

_____

Results

BP _____
Pulse _____
Weight _____
Height _____
BMI _____

Booking Bloods –

Blood Group _____
Hb _____
Platelets _____
TFTs _____
Hep B _____
Hep C _____
Syphilis _____
VZV IgG _____
Rubella IgG _____

Abdomen –

Striations	YES ☐	NO ☐
Linea Nigra	YES ☐	NO ☐
Scars	YES ☐	NO ☐
Scratch Marks	YES ☐	NO ☐