

**PROLACTIN AND TESTOSTERONE LEVELS IN FIRST-TIME FATHERS  
DURING SKIN-TO-SKIN CONTACT WITH THEIR INFANTS SOON AFTER  
BIRTH BY CAESAREAN SECTION**

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## ABBREVIATIONS

AAS: Anabolic androgenic steroids

C/S: Caesarean section

FSH: Follicle-stimulating hormone

GCMS: Gas chromatography-mass spectroscopy

GnRH: Gonadotropin releasing hormone

IRP: International reference preparation

KMC: Kangaroo mother care

LBW: Low birth weight

LH: Luteinising hormone

mg/dl: Milligrams per decilitre

mU/ml: Milliunits per millilitre

ng/l: Nanograms per litre

ng/ml: Nanograms per millilitre

nmol/l: Nanomole per litre

pg/mg: Picograms per milligram

pg/mgCr: Picograms per milligram of creatinine

PrRP: Prolactin-releasing peptide

REC: Research Ethics Committee

REM: Rapid eye movement

rpm: Rotations per minute

SD: Standard deviation

SSC: Skin-to-skin contact

TIDA: Tubero-infundibular dopamine

WHO: World Health Organization

## ABSTRACT

**Introduction and background:** Parental care is vital for the development and survival of offspring. Hormones are known to influence parental care behaviours. Hormonal changes accompanying gestation and delivery have been shown to initiate and maintain maternal care behaviours. Most of the knowledge on hormonal effects on paternal care is from non-human animal studies. Little is known about the effect of hormones on human paternal behaviour.

**Aim:** The aim of the study was to determine the changes in the levels of serum prolactin and testosterone when first-time fathers have skin-to-skin contact (SSC) with their new-born infants immediately after birth by caesarean section (C/S).

### **Hypothesis**

1. That SSC contact between first-time fathers and new-born infants would cause an increase in blood serum prolactin levels.
2. That SSC contact between first-time fathers and new-born infants would cause a decrease in blood serum testosterone levels.

**Research design and setting:** This study was a quasi-experimental study with a one group pre-test and post-test design. The study was conducted at the maternity department of a hospital in Cape Town. The study population consisted of men who were first-time fathers and whose infants were born by C/S. A convenience sampling method was used to select participants. A total of 11 men participated in the study. Six blood samples of 5 ml each were collected from a peripheral vein in the arm of each father, two before the SSC and four during the SSC.

**Data management and analysis:** The serum blood samples were analysed using the Siemens Medical Diagnostics Solutions for prolactin and testosterone assays. The STATISTICA statistical program was used for data analysis. The Wilcoxon matched pairs non-parametric method of analysis was used to analyse the changes in hormone levels.

**Results:** The results of the study did not support the hypothesis. Prolactin levels decreased significantly during the SSC process while testosterone levels were not affected by the SSC process. However, the findings of the study showed an increase in prolactin levels from base levels in the fathers at the time of the birth of the infants.

# CHAPTER ONE: Introduction

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## 1 INTRODUCTION

In the late 19<sup>th</sup> and early 20<sup>th</sup> centuries the care of children in the family was the sole responsibility of the mother in most Western countries. Women were expected to stay at home and had to make sure the children were well nourished and cared for. They had to ensure that the children had a proper upbringing and education by helping with homework from school and teaching their children proper manners and how to behave in society (John, 2000:234). Fathers were regarded as the “breadwinners” of the family and had to make sure that they provided enough money for food and other family needs. They were not expected to spend much time with their children and were mostly considered as disciplinarians in the family (John, 2000:230).

A cross-cultural view of many non-industrialised societies showed child-rearing to be regarded as a woman’s “natural” role. In the early 20<sup>th</sup> century women were more domesticated and their husbands or partners had a more authoritarian position. These roles have gradually shifted over time, as women began to take part in community decisions and assume positions of authority, while men are increasingly seen to participate in the upbringing of their children (Coltrane, 1988:1060).

At the end of World War II there was a dramatic increase in the number of married women participating in the workforce. While mothers worked shorter hours than their husbands and were still responsible for the greater part of the childcare, fathers began to participate in childcare activities (Cabrera et al., 2000:127; Brannen & Nilsen, 2006:346). This brought about a gradual change in the family structure and dynamics, and the role of breadwinner became a responsibility shared by both parents. Today many women work the same number as and sometimes longer hours than their partners; this has required more active involvement of men in the care of their children.

The father-child relationship has been explored by researchers over the last three decades, with a greater number of these studies focusing on the involvement of fathers in the care of their children and the behavioural, cognitive and psychological outcomes of the children. It is evident from the studies that active participation of men in the care of their children from birth through preschool age to adolescence is very important and does not have the same influence on children as the care provided by the mothers (Furstenberg, Morgan & Allison, 1987:699; Yogman, Kindlon & Earls, 1995:58; Black, Dubowitz & Starr, 1999:976; Carlson, 2006:150).

Various factors have been suggested which may influence paternal care both in humans and animals. The economic status and educational levels of fathers have been explored extensively as to how they affect paternal care. In Western societies, men with high incomes and an education level beyond primary school have been reported to be more involved with their children (Bailey, 1993:1033; Yeung et al., 2001:148; Cabrera, Shannon & Tamis-LeMonda, 2007:211). Men who have been married to the same partner for over a decade relate better to their children than men who are divorced, single, or have not been married for a long time (Ahmeduzzaman & Roopnarine, 1992:704; Amato & Sobolewski, 2001:900; Hofferth, 2006:53). Yeung et al. (2001:147) showed that fathers spent more time with their sons when they were teenagers than when they were younger, and more time with their girls when they were younger (i.e. not yet teenagers).

Non-human animal studies have shown that hormones have an impact on paternal care (Wynne-Edwards & Reburn, 2000:464; Wynne-Edwards, 2001:139; Wynne-Edwards & Timonin, 2007:114). However, few studies have been undertaken to explore the effects of hormones on paternal behaviour in humans.

## **1.1 EVOLUTION OF PATERNAL CARE**

Paternal care can be classified as either direct or indirect. Direct paternal care is described as actions by fathers that have an immediate physical effect on their children, have a positive effect on their development and growth, and help to increase their survival rate (for example, carrying,

teaching, grooming, direct feeding, defending, providing direct warmth through cuddling and playing with the young). Indirect paternal care comprises actions carried out by the father that are beneficial to the child but are done without direct contact with the child, such as providing food, shelter and clothing for their young or, in the case of non-humans protecting against predators and defending their territory (Kleiman & Malcolm, 1981:348; Marlowe, 2000:45).

There are several hypotheses that have been used to explain the evolution of paternal care. The “mating effort hypothesis” states that paternal care evolved as a mating strategy which the male used to develop and strengthen a social relationship with the mother. This is seen in non-human primates that live in multi-male, multi-female groups. If a male provided care for the offspring of a female, she would have a preference for him during the mating season (Smuts & Gubernick, 1992:16). Anderson et al. (1999:447) conducted a study in which they interviewed 603 students living and schooling in the township of Gugulethu in Cape Town, South Africa, about the paternal care they received from a biological father or a step-father in terms of time spent with them and monetary investment. The results of the study reported that men provide more care first to their biological offspring, followed by the genetic offspring of their current partner, and least care to children of their previous partner who are not their genetic offspring. This behaviour pattern was interpreted as a means to strengthen the relationship with their current partner.

The “energetic stress hypothesis” states that the energy burden of infant care is greatly reduced when the burden is shared between the parents. According to this hypothesis paternal care evolved as a necessary obligation of the male to help the female carry and groom the neonate during the postpartum period while she is still lactating. By so doing she saves more energy, which is mainly used to breastfeed and in preparation for breeding. In the Black-and-white snub-nose monkey (*Rhinopithecus bieti*) of China it has been observed that paternal care is very high, while maternal care decreases greatly during the months of March to April when temperatures are lowest and food is scarce, compared to the months of July to October when temperatures are high and there is enough food ( $P=0.002$ ) (Xiang, Huo & Xiao, 2010:193).

According to the “paternal investment hypothesis” males are protective over their offspring, and as such will defend them against others and also provide for their needs. A review of behavioural studies on paternity by Busse (1985:874) demonstrated that in multi-male non-human primate groups, males carried their infants almost all the time until they were old enough to defend themselves, in order to prevent infanticide by other males of the group. Human males have been shown to discriminate when it comes to providing for their own offspring rather than those of another man (Anderson et al., 1999:447).

## **1.2 PATERNAL CARE IN PRIMATES**

Of all male mammals approximately 5%, including humans, provide care for their young (Reichard, 2003:5), demonstrating that paternal care is not common among mammals. The involvement of the male in infant care in non-human primates varies between species. There are several species, such as the Patas monkey (*Erythrocebus patas*), the Dusky Leaf baboon (*Trachypithecus obscurus*) and the Capped Langur monkey (*Trachypithecus pileatus*), in which paternal care is almost absent and males do not come into contact with their offspring (Smuts & Gubernick, 1992:4).

In some colonies where a female mates with more than one male, males are able to selectively identify and care for their own offspring and not those of other males. In a study by Buchan et al. (2003:180) 12 out of 15 adult male wild savannah baboons of Kenya (*Papio hamadryas*) studied to determine true paternal care in a multi-male primate society, were observed to provide care to their genetic offspring more than they helped juveniles to whom they were not related ( $P < 0.01$ ). They were also noted to defend their young ones during a dispute more often than they intervened on behalf of the young of other males ( $P < 0.008$ ).

Socially monogamous males have been shown to be more paternally active than socially polyandrous males. Lappan (2008) conducted a cohort study of paternal care in the Siamang ape (*Symphalangus syndactylus*) of Indonesia. This species of non-human primates lives in both

monogamous and polyandrous groups. Three polyandrous and two monogamous groups were chosen and followed over a period of four years (2000-2004), during which infants were born and paternal care was observed. The study showed that monogamous males carried their infants more (15% of the time) than polyandrous males (7% of the time) at 10 to 12 months of age and also at 16 to 18 months of age ( $P < 0.046$ ) (Lappan, 2008:1310).

In some non-human primates infant care is a shared responsibility between all the adult males of the group, even if they are not related to the infant. This is demonstrated in an observational study conducted by Fernandez-Duque, Juarez and Di Fiore (2008) on infant care by adult males other than the biological father in the Owl monkeys (*Aotus azarai*) of Argentina. In this study a solitary adult male is observed to infiltrate a group of monkeys made up of one adult male, one adult female, an infant and an adult son. The solitary adult male was observed to chase away the adult male and carry the three month old infant 67% of the time during their migration (Fernandez-Duque et al., 2008:83).

The involvement of fathers in the care of their offspring varies greatly among human cultures. It ranges from its complete absence, where fathers hardly come in contact with their children (for example, in the Ache fathers of the Paraguayan forest), to great intimacy between fathers and children (for example, the Aka Pygmies of the Western Congo basin in Africa) where men give direct care to their children (Fernandez-Duque, Valeggia & Mendoza, 2009:117). These variations are influenced by factors such as the relationship between the mother and father of the child; the individual characteristics of the child, such as its temperament and gender; public policies such as child support enforcement; and psychological factors such as self-confidence in the fathers (Lamb & Tamis-LeMonda, 2004:11).

### **1.3 PATERNAL CARE AND OFF-SPRING SURVIVAL**

The survival rate of off-spring depends greatly on the care provided by its parents. In some non-human animals the presence of a father improves physical development in the infants. Dudley

(1974) conducted a study to show the effect of paternal care on the growth and development of offspring in California mice (*Peromyscus californicus*), where nine pairs of mice (male and female) were kept in breeding cages and observed for litters. Immediately after the pups were delivered, in five of the nine pairs the young were separated from the father. Every morning for five days all the pups were measured for weight gain and fur growth, after which they were weaned from their mother. Measurements continued until they were 30 days old. Pups raised in the presence of their father were found to have gained more weight than pups raised without their fathers ( $P < 0.01$ ) (Dudley, 1974:158-159). This was probably due to the fact that apart from providing the pup with food, the fathers were also able to provide more warmth with their bodies compared to the warmth provided by the female alone.

In a similar study by Wright and Brown (2002) the effect of paternal care on pup survival in *P. californicus* was explored, where work had to be done to get food. 86 males and females were divided into two groups; in the one group the parents had to run through a wheel which caused an electromagnetic switch to trigger the release of food and the control group was placed in a cage without a wheel. They were further divided into male absent and male present groups. In the male absent group, the males were removed from the cages two days before pup delivery so that the pups were raised only by the female until they were weaned at 24 days of age. In the male present group, the male remained with the female until weaning at 24 days of age. The study terminated after the pups were weaned. More pups were observed to survive in the group where an adult male was present in the cage with the female (77.5%) than in the group with no adult male in the cage with the female (61.2%) ( $P = 0.02$ ). In the wheel group, significantly more pups survived in the presence of a male than in its absence ( $P = 0.04$ ) (Wright & Brown, 2002:46).

In humans, although the father-infant relationship and mother-infant relationship are interconnected, the relationship between the mother and the infant is concerned mostly with feeding and cleaning or grooming, while fathers are more involved in discipline and play activities. As such, the experiences the infant has with the father are different from those he/she has with the

mother. These experiences do not have the same influences on the development of personality from infancy (Lamb, 1977:178; Cabrera et al., 2007:212).

Infant development of preterm babies in terms of weight gain and socialisation has been observed to be positively correlated with the frequency of paternal hospital visits ( $P=0.05$ ) (Levy-Shiff et al., 1990:291). The intellectual and behavioural development of this group of infants when studied longitudinally from birth to three years of age is observed to be greater when fathers are actively involved in their lives from birth while still in the hospital until preschool age (Yogman et al., 1995:58).

Although high income has been positively associated with more paternal care, urban African-American fathers with low income who are active participants in the lives of their children have been shown to contribute positively in the emotional development of their toddlers ( $P<0.001$ ) (Black et al., 1999:972). Positive paternal involvement has also been seen to result in fewer behavioural problems in children. In a study by Amato and Rivera (1999:380) fathers' reports of being caring towards their children were negatively correlated with mother's' reports of home behavioural problems ( $r=-0.22$ ) and school behavioural problems ( $r=-0.16$ ).

A systematic literature review by Sarkadi et al. (2008:153) on 24 publications that were controlled for socio-economic status revealed that active and regular day-to-day participation by fathers in the care of their adolescent children reduced the frequency of delinquency in boys and psychological problems in girls. If the involvement of fathers helps improve the physical, mental and social well-being of their children, this implies that paternal care helps in the reduction of infant and child mortality (Geary, 2000:55).

#### **1.4 EFFECT OF PATERNAL CARE ON THE FAMILY**

The first-time father's involvement in the care of his first child during the first few months of the child's life has been suggested to be very important in determining how he sees himself as a father and also how his partner evaluates him as a parent for both the present child and future children

(Tremblay & Pierce, 2011:99). The involvement of fathers in the care of their offspring is also beneficial for the mother. A study was conducted to measure postnatal depression during the first two months after birth in 119 couples in France. The effects of paternal leave from work and paternal involvement in the care of the new-born were studied (Sejourne et al., 2012). Two self-reporting scales were used: perceived social support by the mothers was assessed using the Multidimensional Scale of Perceived Social Support and depression was assessed using the Edinburgh Postnatal Depression Scale. Questionnaires were completed by the mothers at two to five days postpartum, and again at two month follow up. Paternity leave from work was found not to have an effect on postpartum depression; however, mothers' perception of fathers' participation had a significant impact on postpartum depression ( $P < 0.02$ ) (Sejourne et al., 2012:141). Fathers' involvement in the care of their child is therefore important not only for the development and well-being of the child, but also for that of the family as a whole.

## **1.5 HORMONAL BASIS OF PATERNAL CARE BEHAVIOUR**

Many studies have been conducted to determine the involvement of hormones in parenting. The female brain has been studied using magnetic resonance imaging to observe the brain activity which leads to hormonal release that triggers behaviour towards infant cues such as a hungry cry (Swain, Kim & Ho, 2011:1036). According to Bridges (2008:xxxiv) the involvement of hormones in paternal care has not been studied as extensively as it has in females. It has been suggested that prolactin is the "bonding" hormone, the hormone involved in maternal care. Riddle, Lahr and Bates (1935:730) conducted a study in which repeated injections of prolactin were administered to female virgin rats. This was shown to stimulate maternal instinct and behaviour in the rats to varying degrees. This study was the first conducted on prolactin as a maternal hormone, and paved the way for many more studies, including one conducted by Zarrow, Gandelman and Denenberg (1971:343) which found prolactin to be an important hormone for the expression of maternal behaviour in mammals.

Changes in oestrogen and progesterone levels that are concurrent with gestation and delivery have been established to initiate and maintain maternal behaviour (Insel & Young, 2001:130; Luis et al., 2009:433). Maternal care physiology and behaviours have been studied extensively from as early as the 1930s until the present; however, it was not until the 1990s that emphasis was placed on understanding paternal care. Hormonal involvement in male parenting was first observed in avian species, where males participated fully in nest building, incubation, and feeding of infants (Wingfield & Goldsmith, 1990:94).

Unlike females, males do not get pregnant or breastfeed and are therefore not exposed to the constant hormonal changes which females experience as mothers. However, Wynne-Edwards and Timonin (2007:115) suggest that it may be likely that paternal behaviour and maternal behaviour are stimulated by the same group of hormones. Although this assumption sounds reasonable, the mechanisms involved in paternal care are definitely not triggered by birth and lactation (Wynne-Edwards & Timonin, 2007:115).

Paternal care behaviours have been studied in non-human mammals with specific focus on the neuro-endocrinology of paternal care behaviour (Wynne-Edwards & Reburn, 2000:465). Non-human animal studies have shown a pattern of hormonal change in paternal animals and its absence in non-paternal animals (Wynne-Edwards & Reburn, 2000:464; Storey et al., 2000:79; Roberts et al., 2001:713; Ziegler et al., 2009:436).

In vertebrate animals such as fish, birds and mammals, paternal care behaviour and prolactin have been positively linked (Schradin & Anzenberger, 1999:223; Roberts et al., 2001:713). In some species of non-human primates, prolactin serum levels have been shown to be higher in fathers than in non-fathers, and also to increase in males when they transition from young adult to parents, i.e. when they become fathers for the first time (Schradin et al., 2003:166; Schradin & Anzenberger, 2004:673). This suggests that fathers have higher prolactin levels than non-fathers.

Androgens have been shown to have a negative relation with paternal care. Testosterone is a steroid hormone that has been shown to have an indirect relationship with paternal care. This is

seen in a study by Prudom et al. (2008:604) in which the scent of their infant acutely lowered testosterone levels in fathers while in non-fathers no change in testosterone levels were observed when they were exposed to the scent of a young one (paired t-test=4.87, P=0.005). Some non-human primate males that show direct paternal care, such as carrying the infant for most of the day, have been shown to have very low levels of urinary testosterone (P=0.006) compared to males who hardly had contact with their off-spring (Nunes et al., 2001:74).

Studies across human cultures on paternal care have indicated that men who are married with children tend to have lower levels of testosterone than single men who do not have children (Berg & Wynne-Edwards, 2001:582; Gray, Parkin & Samms-Vaughan, 2007:499). Another study suggests that fathers, as opposed to non-fathers, have low levels of testosterone during the period surrounding their partner's pregnancy and after the birth of their infants (Wynne-Edwards & Reburn, 2000:466).

Apart from testosterone, other steroid hormones known to influence paternal care behaviour are cortisol, oestradiol and progesterone. Oestradiol concentrations have been found to be higher in men who are about to become fathers (68% of the samples) and those whose partners are pregnant than in the controls whose partners are not pregnant (57% of the samples) (P=0.01), while their cortisol levels are lower than in control group (0.30 vs 0.53; P<0.005) (Berg & Wynne-Edwards, 2001:582).

Studies on progesterone and paternal care have shown that progesterone has a negative effect on paternal care in some male species (Trainor et al., 2003:36; Schneider et al., 2003:2951).

Neuropeptide hormones such as vasopressin and oxytocin have also been shown to have an effect on paternal care behaviours. A correlation study between the secretion of vasopressin and paternal care characterised by nest building in Cross-fostered *Peromyscus* mice produced a coefficient (r=0.58, P=0.003), indicating that vasopressin might facilitate paternal care behaviour (Bester-Meredith & Marler, 2003:458). Oxytocin does not have a direct effect on paternal behaviour;

however it facilitates paternal behaviour by being an effective prolactin-releasing factor (Wynne-Edwards & Timonin, 2007:119).

Paternal involvement in the care of the child is not only vital for the development and well-being of the child but for that of the family as a whole. It is influenced by hormonal factors, which are one aspect of many influencing factors. Much of the knowledge on hormonal involvement in male parenting is from non-human animal studies. Very little is known about the role hormones play in the regulation of human paternal care. Knowing more about hormonal involvement in paternal behaviour in humans may be important in understanding the physiology of male parenting.

This study forms part of a larger study being conducted by Dr. Nils Bergman (REC-Ref: 350/2008) on the effect of skin-to-skin care (SSC) of a new-born infant on certain hormone levels in human fathers. Dr. Bergman has specifically studied new-born infants born by normal vaginal delivery. The researcher has therefore included only new-born infants delivered by caesarean section.

## **1.5 PURPOSE OF THE STUDY**

The purpose of this study is to test the hypothesis that when first time fathers have SSC with their infants just after birth, it causes an increase in the secretion of prolactin in the blood and a decrease in testosterone levels. Better understanding of these hormonal changes will help increase knowledge of paternal care behaviours.

### **1.5.1 AIM**

The aim of the study is to determine the changes in the levels of serum prolactin and testosterone when first-time fathers have SSC with their new-born infants after caesarean section (C/S).

### **1.5.2 OBJECTIVES**

- To determine the changes in the level of serum prolactin in fathers over time as SSC is practised between them and their new-born infants.

- To determine the changes in the level of serum testosterone in fathers over time as SSC is practised between them and their new-born infants.

## **1.6 RESEARCH HYPOTHESES**

A hypothesis is a prediction about the relationship between two or more variables (Polit & Beck, 2006). Two research hypotheses were tested in this study.

- That SSC contact between first-time fathers and new-born infants would cause an increase in blood serum prolactin levels; and
- That SSC contact between first-time fathers and new-born infants would cause a decrease in serum blood testosterone levels.

The stated hypotheses above are both directional hypotheses as they specify not only the existence but also the expected direction of the relationship.

## **1.7 SUMMARY**

Human fathers have increasingly become active participants in the care of their children. This care can either be direct or indirect and has been demonstrated to be beneficial for offspring survival. Hormones have been recognised to be one of the factors which influence paternal care. However, little is known about hormonal involvement in paternal care behaviours in humans.

To provide an overview of the rest of this dissertation;

Chapter two presents the review of literature on the production and functions of prolactin and testosterone and their involvement in paternal care. It also investigates the effect of SSC on both the child and parent.

Chapter three discusses the method used in conducting the study, which includes the study design and setting, the study population and sampling method. It also includes the ethical considerations.

Chapter four presents the results obtained from the study in diagrams and tables. It also shows the statistical method used in the analysis of these results.

Chapter five contains a discussion of the results and findings which were presented in chapter four, and uses the literature to justify these findings. It also includes recommendations for further studies.

# CHAPTER TWO: Literature review

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## 2 INTRODUCTION

A review of relevant literature was undertaken. The databases PubMed, CINAHL, Ovid, EBSCO HOST, and Science Direct were searched using the key words: prolactin, testosterone, paternal care behaviour, parental care behaviour and skin-to-skin contact. A search of the Cochrane Libraries was also done. Only English-language articles were reviewed. In addition, a search of the worldwide web using the key terms was conducted for articles related to the study that have not yet been published. Recent literature was sought. A number of old references have been used as these are articles of original work with findings that are relevant to the present study.

Studies in behavioural endocrinology have tested the hypothesis that the endocrine and neural pathway controlling paternal and maternal behaviour is one and the same (Wynne-Edwards & Reburn, 2000:464). If this were so, then the same parental actions and behaviour would be triggered by the same hormones acting at the same neural sites in both males and females (Wynne-Edwards, 2001:139). This demonstrates that paternal behaviour should include all aspects of maternal behaviour except lactation; for example, in the California mouse (*Peromyscus californicus*) males are as actively involved in the care of their young as the females, by building the nest, carrying and licking the pups as well as spending time with them in the nest with the apparent exception of breastfeeding (Schradin & Anzenberger, 1999:226).

Research conducted on male animal species that provide paternal care by nature has provided information to show the part played by hormones in the onset and maintenance of paternal behaviour (Wynne-Edwards & Reburn, 2000:465). Apart from prolactin, other hormones involved in the onset and maintenance of mammalian parental behaviours include oestradiol, progesterone, cortisol, vasopressin and oxytocin (Wynne-Edwards & Timonin, 2007:114).

A review of the synthesis, secretion and function of prolactin was conducted. The production of testosterone and some of its functions were also investigated for the purpose of the study. A review of the available literature regarding how these hormones affect parental behaviour in both male and females was undertaken. Finally literature on SSC also known as kangaroo mother care, was reviewed, including how it affects not only the infant but also the parents.

## **2.1 PROLACTIN SYNTHESIS AND SECRETION**

Prolactin is a polypeptide hormone of the anterior pituitary gland that was originally named for its ability to promote the flow of breast milk as a response to the suckling stimulus of a hungry young mammal (Freeman et al., 2000:1524).

The process of prolactin production occurs mainly in the anterior pituitary gland. The distinct types of cells found in the anterior pituitary gland that synthesize and secrete prolactin are known as lactotrophs (Goffin et al., 2002:48). Fuxe et al. (1977:899) conducted the first study to prove that prolactin was synthesised locally in the brain independent of prolactin synthesis by the pituitary gland. In this study hypophysectomy had no effect on the prolactin-like protein stored in the nerve terminals of the hypothalamic area in the rat brain.

Prolactin is also known to be synthesised and secreted by other organs apart from the anterior pituitary gland and the brain. During pregnancy the placenta has been shown to produce high levels of a hormone known as placental lactogen, which has the same structure and function as prolactin. Placental lactogen sustains the corpus luteum in early pregnancy and stimulates the production of progesterone throughout the pregnancy (Grattan et al., 2008:499). A correlational study by Rosenberg, Maslar and Riddick (1980:681) to demonstrate decidual production of prolactin in late gestation shows very high correlation rate ( $r=0.96$ ,  $p<0.00005$ ) when the ability of the decidua to produce prolactin during pregnancy was correlated with the levels of prolactin in the amniotic fluid. The myometrium of non-pregnant uterus in humans has been demonstrated to

produce prolactin-like proteins which are identical in structure to pituitary prolactin (Gellersen et al., 1991:158).

There is evidence to show that prolactin is also synthesised locally in the breast. Kurtz et al. (1993:1099) conducted a study in which tissue extracts from the mammary glands of pregnant and lactating female Sprague-Dawley and Buffalo rats were cultured and observed for the presence of prolactin messenger ribonucleic acid (mRNA). The epithelial cells of the alveoli and ducts in mammary glands were seen to contain prolactin mRNA. Zinger, McFarland, and Ben-Jonathan (2003) conducted a study to demonstrate the synthesis of prolactin by the different types of breast tissues. Breast tissue was obtained from women undergoing breast reduction or mastectomy and incubated for 10 days. The study showed that both the glandular and adipose tissues in the breast secreted prolactin. In the glandular tissue prolactin secretion increased four-fold by day 10 ( $P < 0.05$ ), while in the adipose tissue prolactin secretion increased 11-fold by day 10 ( $P < 0.05$ ) (Zinger et al., 2003:692).

Immune cells are also a site for prolactin synthesis. A review of literature by Montgomery (2001:665) presents evidence from many studies that demonstrate prolactin synthesis by different immune cells such as lymphocytes, and macrophages from body tissues including the spleen and thymus in humans, mice and rats.

## **2.2 REGULATION OF PROLACTIN SECRETION**

The secretion of prolactin is mainly under the inhibitory control of dopamine. Dopamine is a catecholamine which is synthesised in the hypothalamus by the tubero-in-fundibular dopamine (TIDA) neurons (Ben-Jonathan & Hnasko, 2001:724). Prolactin acts in a short-loop feedback manner to regulate its own secretion by stimulating the TIDA neurons. This means that an acute or chronic increase in blood levels of prolactin triggers the synthesis and release of dopamine by the TIDA neurons. Dopamine released is then transported in the blood through the hypophyseal portal vessels to the pituitary gland, where it activates the D2 (type 2 dopamine) receptors on the pituitary

lactotrophs to inhibit prolactin secretion. Hypoprolactinaemia, or a decrease in levels of blood prolactin, results in the suppression of dopamine secretion by the hypothalamus (Grattan & Kokay, 2008:753).

Regulation of prolactin secretion has also been shown to take place by other mechanisms. Grattan and Averill (1991:401) demonstrated that the placenta not only produces prolactin but also regulates its release at different times during pregnancy. In early pregnancy progesterone produced by the placenta helps stimulate prolactin secretion, while in late pregnancy it impedes the secretion of prolactin. The ovaries have also been shown to regulate secretion of prolactin. De Paul et al. (1997:287) reported that rats whose ovaries have been removed experienced a decrease in lactotrophs and decreased prolactin secretion. Oestrogen, a hormone secreted by the ovaries, was administered to these rats and it was observed that prolactin secretion increased significantly ( $P < 0.05$ ).

Although the non-pregnant uterus has been shown to be a source of prolactin production, there are data that suggest that the epithelial layer of the uterus produces certain elements that act as inhibitory factors to the secretion of pituitary prolactin (Gorospe & Freeman, 1985:1559). Freeman et al. (2000:1582) conducted a systematic literature review which provides evidence that adrenalectomy (removal of the adrenal gland) enhances prolactin secretion.

## **2.3 PATTERN OF PROLACTIN SECRETION**

The classic pattern of prolactin secretion is caused by the action of an infant suckling on the nipple. This action produces a signal in the afferent somatosensory neural pathway which is relayed to the hypothalamus and then the pituitary gland, inducing secretion of prolactin. When suckling stops, the secretion of prolactin also stops as the suckling stimulus ends. (Freeman et al., 2000:1537). There are, however, other patterns of prolactin secretion.

### **2.3.1 CIRCADIAN PATTERN**

Prolactin has been shown by some studies to have a circadian pattern of secretion, with high concentrations occurring during the night and during periods of sleep. In a study by Nokin et al. (1972:561) seven adult men, 12 non-pregnant women and three pregnant women in their last month of pregnancy provided blood samples at four hour intervals for a period of 24 hours in which prolactin levels were measured. The study revealed that non-pregnant women had higher prolactin levels at 01h00 (mean 500 mU/ml) and 05h00 (mean 504 mU/ml) than at 09h00 (mean 332 mU/ml), 13h00 (mean 273 mU/ml), 17h00 (mean 299 mU/ml) and 21h00 (mean 331 mU/ml) ( $P>0.05$ ). Mean prolactin levels in men were also raised at 01h00 (270 mU/ml) and 05h00 (354 mU/ml) compared to values at 09h00 (209 mU/ml), 13h00 (222 mU/ml), 17h00 (222 mU/ml) and 21h00 (205 mU/ml), ( $P>0.001$ ). No circadian change was observed in the pregnant women.

### **2.3.2 SEASONAL PATTERN**

Seasonal patterns of prolactin secretion are observed in mammals that breed seasonally, with highest plasma prolactin concentrations in summer and spring, and lowest concentrations in autumn and winter. This seasonal change has been suggested to be due to the hormonal susceptibility in these animals to changes in daylight time, as winter and autumn have longer nights than summer and spring (Curlewis, 1999:1). Lincoln (1990) demonstrated seasonal patterns of prolactin secretion in the ram. Groups of four to seven rams were studied for two annual cycles and blood samples were collected every six months; the results showed an 18 to 66 fold increase in prolactin concentrations from the winter periods between November to December, with maximum values in summer (May to June) ( $P<0.001$ ) (Lincoln, 1990:287).

### **2.3.3 SLEEP/WAKEFUL STATE PATTERN**

The relationship between wakeful state, sleep and prolactin was studied by Parker, Rossman and Van der Laan (1974:646). Blood samples were drawn from 14 adult male participants during 58 nights of monitored sleep. Results demonstrated prolactin serum levels being lowest during wakeful state and rapid eye movement (REM) sleep, with values peaking during the non-REM sleep cycle. Another study demonstrated prolactin secretory rate to be lowest at the time of REM

sleep onset (Spiegel et al., 1994:20). Lincoln and Clarke (1994:251) provided evidence that melatonin, a hormone secreted by the pineal gland in the absence of light, bypasses the hypothalamus and acts directly on the pituitary gland to secrete prolactin, hence the high levels of prolactin during night time.

## **2.4 FUNCTIONS OF PROLACTIN**

Prolactin is a hormone involved in many biological actions in the human body. Two of the most relevant actions are discussed briefly below.

### **2.4.1 REPRODUCTIVE ROLE OF PROLACTIN**

Prolactin is an important factor in the production and secretion of breast milk. It also plays an important role in the maintenance and function of other organs of reproduction in both the female and the male.

#### ***2.4.1.1 FEMALE REPRODUCTION***

Studies on rodents have shown that prolactin is important in maintaining the structure and function of the corpus luteum in the female during the first few weeks following fertilization (Freeman et al., 2000: 1535), maintaining pregnancy to full term and preventing miscarriages (Bachelot & Binart, 2007: 363).

Prolactin together with other growth hormones such as progesterone and growth hormone has been demonstrated to support the proliferation of the mammary glands during pregnancy. It has also been demonstrated to sustain the production and accumulation of breast milk in preparation for breastfeeding (Neville et al., 2002: 53).

#### ***2.4.1.2 MALE REPRODUCTION***

Prolactin and its receptors have been found in the epithelial cells of the human and rat prostate and have been shown to influence the morphology and synthesis of the prostate gland (Nevalainen et al., 1997:624). A study by Steger et al. (1998:3694) on the effect of prolactin on male reproduction

in mice suggested that it plays a part in the growth and differentiation of accessory organs such as the prostate and seminal vesicles, but is not important in maintaining the fertility of males. Hence prolactin plays a minimal role in male reproduction.

## **2.4.2 HOMEOSTATIC ROLE OF PROLACTIN**

Apart from its role in reproduction, prolactin also plays a role in the maintenance of balance in the internal environment in the body.

### ***2.4.2.1 IMMUNE RESPONSE***

Gala (1991:513), in a review of studies on prolactin regulation of the immune system, showed that animals in which the hypothalamus was removed had deficient immune function which was corrected by treatment with prolactin. This review also showed a number of clinical trials in which prolactin and growth hormone stimulates the thymus gland to secrete thymulin, a hormone which helps improve immune function in humans (Gala, 1991:518).

### ***2.4.2.2 OSMOREGULATION***

Prolactin is an important hormone involved in the transportation of electrolytes across cells. Bussieres et al. (1987:182) evaluated the effect of prolactin on the regulation of water and electrolytes by the kidneys. Rats used in the study were administered prolactin while the controls were injected with bromocriptine (a prolactin antagonist). Three hours later a decrease of about 80% in the movement of sodium and potassium across the cells in the kidneys was observed in the rats which had been given bromocriptine ( $P < 0.001$ ), while in the rats injected with prolactin, sodium and potassium movement increased and persisted for up to 6 hours later ( $P < 0.001$ ). Correlation of electrolyte activity and prolactin plasma levels produced a correlation coefficient of 0.89. Administrations of prolactin in vitamin D-deficient male mice has been observed to induce the absorption of calcium in the small intestine to increase up to four-fold ( $P < 0.05$ ) (Ajibade et al., 2010:2977).

### **2.4.2.3 BODY WEIGHT**

It has been hypothesised that prolactin plays a role in the regulation of body weight. In a study conducted by Lawrence, Ellacott, and Luckman (2002:261) prolactin-releasing peptide (PrRP) was administered to adult male rats that had been starved for 24 hours. Food was then given to these rats and the amount of food consumed was measured. The results showed a significant decrease in food intake and body weight (-1+-2g) when compared to the control group (4+-1g) ( $P < 0.05$ ). Similarly, Gu et al. (2004:98) conducted a study using mice in which the PrRP gene had been deleted so that they did not secrete prolactin. They observed that at 16 weeks of age in the male and 26 weeks of age in the female, these mice had a higher body weight when compared to the controls. The mice were also observed to consume more food per week (22% more) than the control group ( $P < 0.01$ ) (Gu et al., 2004:100).

Prolactin may have important functions in the body, but it has also been shown to be involved in the formation of tumours. Clevenger, Furth, Hankinson and Schuler (2003:2) conducted a literature review to demonstrate the involvement of prolactin in breast cancer. A study by Dagvadorj et al. (2007:3029) concluded that prolactin promotes cell growth and viability of cancerous prostate cells.

## **2.5 PROLACTIN AND PATERNAL CARE BEHAVIOURS**

Of all hormones playing a role in parenting, prolactin is regarded as one of those with an important role in the expression of parenting in both males and females (Ziegler et al., 2000:111). An increase in levels of prolactin in the female during the process of child birth may seem to be a result of the constant secretion of high prolactin levels by the placenta during pregnancy. This increase in the secretion of prolactin is maintained after delivery by the suckling stimulus of the child (Grattan et al., 2008:499). It has been suggested that male prolactin levels probably respond to different mechanisms than those seen in the female (Ziegler et al., 2000:118).

Prolactin plays an important role in maternal behaviour. It has been shown to stimulate the onset of maternal behaviour in female rats that have not been pregnant before (Slotnick, Carpenter &

Fusco, 1973:53; Bridges et al., 1985:782). Bridges et al. (1990) conducted a study in which female nulliparous rats whose ovaries had been removed and which had been injected with bromocriptin to prevent endogenous production of prolactin were administered synthetic prolactin and observed for maternal behaviour towards fostered young rats. It was observed that prolactin administered centrally into the lateral ventricle stimulated maternal behaviour, unlike prolactin which was administered subcutaneously ( $P < 0.05$ ) (Bridges et al., 1990:8006).

### **2.5.1 PROLACTIN AND PATERNAL CARE IN NON-HUMAN PRIMATES**

Correlational studies have been conducted regarding prolactin and paternal care behaviours. The majority of these studies have been conducted on non-human animals including birds, rats and monkeys. These studies have led to questions of whether prolactin plays a causal role in stimulating and maintaining infant care in males.

Brown et al. (1995) examined the hormonal changes which the male Mongolian gerbil mouse (*Meriones unguiculatus*) undergoes throughout the reproductive cycle of a female mate. Couples were kept in cages and the hormonal levels of the males were examined from the time of mating until the pups were ready for weaning. The control group was made up of adult males who were not placed in a cage with females. Paternal activity was seen in the form of shredding paper and nest building while the female was still pregnant, crouching over a pup and licking when the pups were born. The study revealed prolactin levels to rise continuously in the males after mating until 20 days after the pup was born, unlike in the controls ( $P < 0.05$ ) (Brown et al., 1995:478). This led to the conclusion that paternal behaviour in these males is initiated by neuro-endocrine changes induced through cohabitation with the female.

Fathers of some species of New World Monkeys (monkeys that belong to the genera *Callithrix*; *Saguinus*; *Callimico*; *Callicebus*) experience an increase in prolactin levels after the birth of their infants. Males of the common marmosets (*Callitrix jacchus*) that have no paternal experience exhibit elevated serum prolactin levels after infant carrying (Roberts et al., 2001). In this study, seven adult males who had no prior paternal experience were exposed to the cry, smell and sight of infants and their responsiveness in terms of infant retrieval and carrying was measured. Blood

samples were collected before and after the infant carrying. Their mean prolactin level after carrying infants was  $18.8 \pm 3.95$  ng/ml compared to their mean prolactin level of  $6.53 \pm 0.97$  ng/ml before the carrying. The prolactin level of monkeys that never carried an infant was  $6.9 \pm 1.16$  ng/ml (Roberts et al., 2001:717).

In a similar study by Schradin et al. (2003), nine fathers of the *Callicebus cupreus* species, seven fathers of the *Callithrix jacchus* species, and seven of the *Callimico goeldii* species were used to determine if prolactin levels increase in males after infant birth. They were housed together with a pregnant female. Early-morning urine samples were collected every day from three weeks prior to the birth of an infant until three weeks post-birth. Prolactin mean values in *C. cupreus* were 33pg/mg three weeks prior to infant birth, and 41 pg/mg in weeks one to three after birth (n=6). *C. jacchus* fathers had raised prolactin levels (mean value 102 pg/mg) after the birth of their infants compared to a mean of 76 pg/mg prior to the birth of their young (n=7). In the *C. goeldii*, mean prolactin levels were 68 pg/mg before birth increasing to 104 pg/mg after the birth of the infant. However, in all three species, no correlation was found between prolactin and infant carrying during the first three weeks after birth of the infant (Schradin et al., 2003:170). Hence, although prolactin levels are raised after infant birth in the New World Monkeys, not all species show increases in prolactin levels during infant carrying.

Progressive rises in prolactin levels have been observed in the *C. jacchus* as they mature from adults to parents (Schradin & Anzenberger, 2004:673). The study showed mean urine prolactin levels of 3pg/mgCr when they become adult males, 40 pg/mgCr before the birth of their first infants and 80 pg/mgCr one day after the birth of the infant, indicating a progressive rise in prolactin as these monkeys matured from adults to parents. This value is seen to drop to 35 pg/mgCr 10 weeks post birth. Multiple comparisons showed high significance when prolactin levels immediately after birth were compared with levels preceding birth of their infant ( $P < 0.01$ ), and when compared to prolactin levels as adult sons ( $P < 0.05$ ). However correlation between prolactin and age in these males was insignificant. This implies that the progressive increase is not as a result of age but perhaps due to the presence of a female just before birth and of an infant after birth.

In the socially monogamous cotton-top tamarin (*Saguinus oedipus*) males that have previously fathered an infant demonstrate a greater increase in levels of prolactin both before and after their current mate's pregnancy when compared to adult non-father males (U=49, P=0.002). It has also been shown that experienced males carry infants more during the first five days postpartum than inexperienced males (U= 11.5, P=0.05) (Ziegler & Snowdon, 2000:162). Ziegler, Wegner and Snowdon (1996:219) demonstrated that fathers of this species have significantly higher prolactin levels than non-fathers (P=0.005), and that experienced fathers have significantly higher mean prolactin levels than first-time fathers (P<0.001). The results also demonstrated first-time fathers to have prolactin levels that are significantly higher after the birth of their infant (mean prolactin value 0.6 ng/mgCr) than they had when paired with their non-pregnant mates (mean prolactin value 0.2 ng/mgCr), (P=0.003) (Ziegler et al., 1996:291).

Contrary to these results a study by Schradin and Pillay (2004:48) on prolactin levels in the paternal striped mouse of Southern Africa (*Rhabdomy pumilio*) showed no increase in prolactin levels in fathers compared to non-fathers (P> 0.1; U=204). However, the study showed that experienced fathers had significantly higher levels of prolactin (22.25±3.3 ng/ml; n=11) compared to inexperienced fathers (16.1±2.2 ng/ml) (Schradin & Pillay, 2004:47).

Although these studies in non-human mammals show that prolactin may stimulate and maintain paternal care behaviour, other studies show the contrary. Almond, Brown and Keverne (2006:676) conducted a study in which prolactin levels were suppressed to insignificant levels of 0.005 ng/ml using cabergoline (a prolactin suppressant) in the paternally experienced common marmosets (*C. jacchus*) (n=5). It was observed that the amount of infant carrying, feeding, retrieval and grooming was not affected by low prolactin levels. In a similar study, prolactin was suppressed by injecting 300 micrograms of bromocriptine subcutaneously three days prior to the birth of pups in the Djungarian hamster (*P. campbelli*) (n=22). This was observed not to have an effect on the retrieval and carrying of displaced pups during the 3 days of paternal behaviour observation in seventeen of the eighteen males used in the study (P<0.002) (Brooks, Vella & Wynne-Edwards, 2005:362).

## 2.5.2 PROLACTIN AND PATERNAL CARE IN HUMANS

Studies on humans suggest that fathers have higher prolactin levels than non-fathers. A study conducted to determine hormonal changes in expectant fathers reported that first-time fathers showed an increase in serum prolactin levels just before infant birth and when exposed to their infant's cry immediately after birth, when compared to men whose partners were still in the first trimester of pregnancy ( $P=0.05$ ). The study also showed that experienced fathers have a higher increase in prolactin on hearing an infant cry compared to first-time fathers ( $P=0.06$ ) (Storey et al., 2000:84). Gettler et al. (2012:3629) have also shown that human fathers have higher prolactin levels than non-fathers ( $P=0.006$ ), and that fathers of younger children have higher prolactin levels than fathers of older children ( $P=0.054$ ).

Fleming et al. (2002) conducted a study to determine the hormonal changes and emotional response of fathers and non-fathers to infant cues. Emotional response was measured by a questionnaire describing responses such as disturbed, distressed, annoyed, happy, worried and sympathetic. A finger-prick blood sample and saliva were collected before the stimuli, within one minute after the stimuli and 20 minutes after the stimuli. The results showed fathers to be more sympathetic than non-fathers towards infant cues ( $P<0.05$ ), and that prolactin levels increased more in fathers than non-fathers with experienced fathers having a higher percentage increase ( $P<0.004$ ) (Fleming et al., 2002:406).

Although prolactin is seen to increase initially when men are exposed to infant cues, it has been shown to decline as contact time between the father and child increases. Gettler et al. (2011:599) conducted a study in the Philippines which showed prolactin to decrease in fathers after they played with their children for a period of 30 minutes. The study also revealed that first-time fathers had a greater decline in prolactin compared to fathers who had more than one child ( $P<0.05$ ), and that prolactin levels decreased more in men who felt their wives had a positive impression about them as care givers compared to men who felt their wives evaluated them less positively ( $P<0.01$ ).

Storey et al. (2011:356) recruited 12 fathers who volunteered to participate in a similar study, which was conducted when their child was between 20 and 26 months old. They were tested on two

occasions, on a 'with-child' day when they spent the whole day with the child before the test were done, and on a 'without-child' day when they spent several hours away from the child before the test were done. Blood samples were collected just before a 30 minutes interaction with their child and immediately after the interaction. It was observed that prolactin levels declined significantly from the baseline values after a period of 30 minutes of play activity with their toddlers ( $P < 0.001$ ) on both days when they spent the entire day with their child or away from their child.

In summary, prolactin levels have been demonstrated to increase more in fathers than in non-fathers, and increase in prolactin levels could drop as contact time between the father and the child increases.

## **2.6 TESTOSTERONE SYNTHESIS**

Testosterone is a 9-carbon steroid and is the predominant androgen in most mammalian male species. Approximately 95% (3 - 10 mg/dl) of the testosterone found in most mammalian male species is obtained from testicular secretions (Glickman et al., 1992:451).

The production of testosterone in the testes is by a heterogenous group of cells which include Leydig cells, Leydig cell precursors and immature Leydig cells. The secretion of testosterone by these cells is under the control of luteinising hormone (LH), a hormone secreted by the pituitary gland (Bhasin, 2008:645). A study by Mendis-Handagama et al. (1998:67) to demonstrate the effect of LH on testosterone production showed that adult male rats treated for two weeks with LH had an increase in Leydig cell volume of two fold per testis ( $P < 0.05$ ), and an increase of testosterone secretion of up to six fold per Leydig cell ( $P < 0.05$ ) as compared to the control group. Leydig cell synthesis of testosterone is a complex pathway which involves various enzymes that break down cholesterol into progesterone and finally to testosterone, and is modulated by certain growth factors such as gonadotropin-releasing hormone (GnRH) and vasopressin. Cytokines within the seminiferous tubules of the testes also modulate testosterone production by the Leydig cells (Bhasin & Jameson, 2008).

## **2.7 TESTOSTERONE SECRETION**

Bhasin (2008:646) states that “Testosterone secretion by the Leydig cells is regulated by feed-forward and feedback mechanism that operates within the hypothalamic-pituitary-gonadal axis” The release of GnRH by the hypothalamus stimulates the anterior pituitary to release LH, which in turn stimulates the synthesis of testosterone by the Leydig cells in the testes. Very high testosterone levels in the blood inhibit the release of LH by both decreasing hypothalamic GnRH secretion and by decreasing pituitary responsiveness to GnRH (Matsumoto & Bremner, 1984:612). Foresta et al. (1997:3041) reported that an increase in the concentration of LH in the peripheral blood preceded a rise in the concentration of testosterone in the spermatic veins by about 120 minutes ( $P < 0.002$ ), demonstrating a feed-forward mechanism, and that testosterone increases in the spermatic veins preceded a drop in the concentration of serum LH by about 40 minutes ( $P < 0.05$ ), showing a feedback mechanism.

## **2.8 BIOLOGICAL ACTIONS OF TESTOSTERONE**

Testosterone is a hormone known to increase muscle mass, especially in men. Data from a study conducted to determine the effect of testosterone on muscle activity demonstrated that testosterone increases the voluntary action and power of leg muscles, but does not improve tired muscles in young adult males. This effect depends on the dose and concentrations administered (Storer et al., 2003:1482). Anabolic Androgenic Steroids (AAS) are synthetic derivatives of testosterone that can be administered to individuals orally, parenterally or topically. They function mostly to maximise anabolic effects while minimising androgenic effects (Evans, 2004:535). AAS are mostly used by athletes as they are considered as performance-enhancing drugs and increase the motivation to compete (Wood & Stanton, 2012:149). Clark and Henderson (2003:419) however, conducted a review of behavioural and physiological responses to AAS which showed that most often human males that use AAS report a decrease in sperm production.

Testosterone has been shown to improve memory and learning. Leonard and Winsauer (2011:545) reviewed several studies in which a male rat injected with testosterone spent more time exploring a new object, odour or taste than an old one, while a male that had not been injected with testosterone spent almost the same amount of time on both a new object and an old object, indicating that there is probably no memory of the old object. Several studies on maze retrieval by rodents were also reviewed, and the conclusion drawn was that testosterone enhances various memory and learning tasks in the male either directly or indirectly through its derivatives such as oestradiol.

### **2.8.1 REPRODUCTIVE ROLE OF TESTOSTERONE**

The formation of the male phenotype following the weeks after conception is mainly controlled by the activity of testosterone. It is responsible for the growth of facial and pubic hair, deepening of the voice and broadening of the chest in boys, changing them into men at puberty (Evans, 2004: 534). Testosterone is the most important androgen responsible for the normal maintenance of sperm production (spermatogenesis) (Lacy & Pettitt, 1970:87).

### **2.8.2 TESTOSTERONE AND BREEDING**

In non-human vertebrates higher testosterone levels have been shown to increase mating strategies in the male. In the Soay Sheep of the Hirta region of Scotland, increased testosterone levels in the male have been associated with increased mate-seeking behaviour and aggression towards other males during the mating season ( $P < 0.008$ ) (Preston et al., 2012:301). A positive link between male-male dominance interactions (in territory defence, courtship and mate guarding) and testosterone during the breeding season has been observed in some Mexican bird species (Vleck & Brown, 1999:943). Contrary to this, Bonobo monkeys of the Democratic Republic of Congo which are aggressive in the presence of a female, show low levels of testosterone ( $P = 0.01$ ) (Surbeck et al., 2012:663). In the wild chimpanzee of Uganda individual rates of aggression in the presence of a fertile female were not strongly correlated with urinary levels of testosterone ( $r = 0.21$ ,  $P = 0.13$ ) (Muller & Wrangham, 2004:120). In human males, increased libido is positively related to elevated testosterone levels (Wang, Swerdloff et al., 2000:2839).

## 2.9 TESTOSTERONE AND PATERNAL CARE BEHAVIOURS

Testosterone has been shown to have no effect on paternal care in some non-human mammalian species. In a study by Hume and Wynne-Edwards (2005: 306), paternal behaviour in the biparental dwarf hamster (*Phodopus cambelli*), which was characterised by making contact with the pup, licking of the pup, picking up the pup and retrieving a displaced pup, did not change after animals had been castrated, causing a reduction in testosterone concentrations below the lower limit of 0.2 ng/ml. This showed that paternal behaviour was independent of the changes in testosterone levels. Lower testosterone levels have been observed in males of these same species that never retrieved a displaced pup during a series of three tests conducted over a three-day period compared to levels of testosterone in males that showed paternal behaviour by retrieving a pup at least once (Schum & Wynne-Edwards, 2005:415).

In other non-human mammalian species, testosterone has been shown to promote paternal care. In Prairie voles (*Microtus ochrogaster*) castration has been shown to reduce paternal responsiveness towards its young, while castrated *M. ochrogaster* that have been treated with testosterone demonstrate paternal behaviour such as liking and picking up displaced pups (Wang & De Vries, 1993:156). Trainor and Marler (2002:825) conducted a study in which testosterone was converted to oestradiol through the process of aromatisation and administered to castrated male California mice. These mice were observed to show a significantly high level of paternal care through huddling and grooming when compared with the control group that received a placebo ( $P < 0.001$ ). This implies that testosterone promotes paternal care indirectly through oestradiol.

In a similar study by Luis et al. (2009:437), 14 male Volcano mice (*Neotomodon alstoni*) were observed for 21 days post-delivery for paternal behaviour which was characterised by huddling, cuddling, sniffing and carrying of a pup. Blood samples were collected at day 10 after mating, day five post-partum, and day 20 post-partum. A positive correlation between levels of testosterone and the amount of paternal care provided by the male Volcano mouse was observed at day five post-partum ( $r = 0.73$ ,  $P < 0.5$ ) There was no correlation between testosterone levels and care at day twenty postpartum ( $r = -0.3$ ,  $P < 0.05$ ). The male Mongolian gerbil has been shown to maintain high

levels of testosterone during the mate's pregnancy which is probably associated with aggression towards other males. These levels increase postpartum and have a positive correlation with the time spent with the pup ( $r=0.68$ ,  $P<0.05$ ) (Juana et al., 2010:272).

Other studies suggest a link between lower testosterone levels and paternal care. Plasma testosterone levels have been observed to decrease significantly in two different species of the male songbird from the pre-nesting stage to the nestling stage when they build nests in preparation for the female to lay eggs ( $P=0.024$ ) (Van Roo, Ketterson & Sharp, 2003:432). Prudom et al. (2008:604) conducted a study which demonstrated that testosterone levels dropped in experienced male marmoset fathers when they were exposed to the scent of their infants compared to when they were exposed to a control scent (paired  $t$ -test= $4.87$ ,  $P=0.005$ ). It also showed that in non-fathers there was no significant change in serum testosterone when exposed to an infant's scent and the control scent (paired  $t$ -test=  $-0.56$ ,  $P=0.61$ ). In other words, a high level of testosterone inhibits paternal care in some non-human animals, as demonstrated in a study by Van Roo (2004). In his study paternal care characterised by egg incubation, nest cleaning and feeding of the young in the monogamous blue-headed vireos males, was observed to decrease significantly when they were treated with testosterone (Van Roo, 2004:678).

### **2.9.1 TESTOSTERONE AND PATERNAL CARE IN HUMANS**

Low testosterone levels have been observed in men who are becoming fathers. Berg and Wynne-Edwards (2001:586) conducted a study in which men living with a pregnant partner were found to have significantly lower testosterone levels than men whose partners were not pregnant ( $P=0.005$ ). Fathers living with their children have demonstrated lower levels of testosterone than non-fathers ( $P<0.05$ ) (Gray et al., 2007:502). Low testosterone levels have also been observed in men of two neighbouring Tanzanian groups (the Hadza foragers and the Datoga pastoralists) who show high levels of paternal care than in non-fathers in both morning and evening urine samples. (Morning -fathers:  $124\pm 13.6$  pmol,  $n=10$ ; non-fathers:  $176\pm 17.7$  pmol,  $n=15$ ;  $z=-0.2164$ ,  $p=0.03$ ; Evening -fathers:  $83\pm 8.83$  pmol,  $n= 10$ ; non-fathers:  $157\pm 16.3$  pmol,  $n=15$ ;  $z=-2.691$ ,  $p=0.007$ ) (Muller et al., 2009:350).

Among Swahili men in Kenya correlation analysis shows that married men with younger children have lower testosterone levels than those with older children (Gray, 2003:282), while in the Philippines fathers with infants less than one year old are reported to have testosterone levels 10 mg/dl lower than fathers of older children (Kuzawa et al., 2009:434), and fathers of neonates experience a greater decline in testosterone levels than fathers of toddlers ( $P < 0.003$ ) (Gettler et al., 2011:16195). This may be due to the fact that younger children are more dependent on parents than older ones.

In a study to determine whether fatherhood decreases testosterone in men, 624 men with a mean age of 26 years were recruited in the Philippines as single young adults and followed up once they had become paired and had their first child (four years after initial recruitment). Two salivary testosterone samples were collected on enrolment (evening and morning samples) and two samples four years later. Men who were newly married and had their first child demonstrated a greater decrease in serum testosterone levels from baseline values in both morning and evening samples than unmarried men who had no children ( $P < 0.001$ ). Men who were married but had no child showed a decrease in testosterone levels that was not significantly different from that of unmarried men ( $P = 0.167$ ) (Gettler et al., 2011: 16195).

A study in Beijing, China also demonstrated that married men with children had significantly lower testosterone levels than married non-fathers or unmarried men ( $P < 0.0001$ ) (Gray, Yang & Pope, 2006:335). The decrease in testosterone levels in human males as they transition from being non-fathers to becoming fathers could be seen as a strategy to allow males to mediate trade-off between mating efforts and paternal efforts, since high levels of testosterone are required for mating (Goymann, Landys & Wingfield, 2007:463).

## **2.10 KANGAROO MOTHER CARE**

Kangaroo mother care (KMC), also known as Skin-to-Skin Care (SSC), was introduced in 1978 by Drs Martinez and Reys in Bogota, Columbia, and first implemented at the maternal and child unit of Bogota Hospital. It was used for early hospital discharge of low birth weight (LBW) infants due to the critical shortage of medical equipment and resources, cross-infections, high neonatal mortality rates and child abandonment by mothers who were discharged but had sick infants requiring them to stay in hospital (Venancio & de Almeida, 2005:173).

“Kangaroo mother care is the positioning of a near naked infant at or between the mother’s breasts so that there is a skin-to-skin contact between the two” (Roberts, Paynter, & McEwan, 2000:31). The term “Kangaroo Mother Care” comes from the fact that the practice is similar to that of marsupials. Marsupials’ infants are born prematurely and are kept in their mothers’ pouch permanently where they receive feeding and warmth until they are mature enough to be able to survive outside (Tessier et al., 1998:1). KMC can be either continuous or intermittent. In continuous KMC, the mother and infant are in the kangaroo position 24 hours a day while intermittent KMC is for short periods a few times per day (Nygqvist et al., 2010:821).

There are three main elements that form part of KMC: regulation of body temperature through the closeness of the SSC between the mother and the infant; exclusive breastfeeding or other form of suitable feeding method; and prompt determination of and reaction to any problems in the infant. It is advocated that the infant is colonised by the mother’s commensal organisms, which help to minimise the possibility of the infant developing nosocomial infections (Tessier et al., 1998:3).

World Health Organization (WHO) (2003) guidelines recommend that KMC is initiated in hospitals and can be continued at home by the mothers with adequate support and follow-up by hospital staff. This enables stable preterm babies to be discharged early. The WHO also recommends that KMC should be started soon after birth, and should be continuous and prolonged for as long as possible with exclusive breastfeeding in ideal situations (WHO, 2003:2). KMC has been shown to be feasible, cost effective and readily acceptable by hospital staff and mothers in developing countries (Cattaneo et al., 1998:979).

### **2.10.1 KANGAROO POSITION**

Nyqvist et al. (2010:824) define the kangaroo position as that where the infant is placed in SSC on the chest of the mother between her breasts. The infant is securely placed either vertically or semi-reclined with flexed arms and legs in a froglike position, with the head turned sideways and upright (not flexed or extended). It is vital to ensure the patency of the infant's airway. The infant may wear a cap to avoid hypothermia and a diaper to avoid wetting the mother.

### **2.12 ADVANTAGES OF KMC TO NEWBORN INFANTS**

Continuous KMC immediately after birth has been shown to reduce the rate of neonatal and infant mortality, especially in low-income countries, through the effective means of timely temperature stabilisation and early establishment of breast-feeding (Conde-Agudelo, Diaz-Rossello & Belizan, 2003:5; Lawn et al., 2010:144). Preterm infants nursed in incubators are more likely to experience hypothermia and respiratory distress than those nursed in KMC. They are also likely to gain less weight ( $P < 0.001$ ) as the frequency and rate of breastfeeding is less than if they are in SSC with their mothers (Cattaneo et al., 1998:979; Bergman, Linley & Fawcus, 2004:783; Suman Rao, Udani & Nanavati, 2008:17; Mori et al., 2010:161).

KMC has been shown to improve the survival rate of preterm babies and LBW infants. Lincetto, Nazir and Cattaneo (2000:294) conducted a study which demonstrated that 73% of preterm infants who were put into KMC immediately after birth survived, compared with 38% who were not nursed in KMC but in an incubator ( $P < 0.01$ ). The frequency of nosocomial infections in LBW infants has been shown to be higher in infants nursed in incubators (6.8%) than in those nursed in KMC (3.4%) (Charpak et al., 2001:1074).

Apart from preventing hypothermia and respiratory illness in the preterm infant, it has been hypothesised that KMC has a positive impact on the physiological development of preterm and LBW infants. A study was conducted to compare preterm development outcome in both SSC and traditional care (incubator care). Univariate tests showed that after continuous practice of SSC

while in hospital, infants scored higher on both the mental and psychomotor development indices of the Bayley-II (instrument to measure development) at 6 months corrected age (mean 85.14, standard deviation (SD) 13.68) than those in the traditional care group (mean 77.91, SD13.68) (Feldman et al., 2002:21).

In a literature review exploring KMC as a developmental intervention affecting growth and development in preterm infants, Dodd (2005:224) reported that sensorimotor stimulation promotes growth and development in the preterm. Handling and stimulating actions like rocking, loving touch and massaging by the parents are gentle sensorimotor stimulations that enhance the development of the preterm baby. Infants receiving KMC have been observed to have a greater daily weight gain than those not in KMC.

Term infants placed into SSC with their mothers after normal birth have been observed to spend more time in restful sleep state ( $F [1, 45] = 5.90; P=0.019$ ) and less time in fussing, crying and alert states ( $F [1, 45] = 6.75; P=0.019$ ) when compared with the control infants who were taken to the nursery after delivery without SSC (Ferber & Makhoul, 2004:860). Full term infants with cardiac pathologies have been found to have lower mean heart rates postoperatively during SSC provided after extubation, suggesting that SSC decreases postoperative stress and discomfort in the baby (Gazzolo, Masetti & Mali, 2000:729).

When placed in SSC with their fathers, term infants born by C/S stopped crying and became drowsy and sleepy within 60 minutes of being born, while those placed in cots reached a drowsy state after 110 minutes ( $P=0.001$ ) (Erlandsson et al., 2007:105). KMC might therefore be a useful intervention in the reduction of stress associated with the birth process in the term infant.

In a study to determine the effects of KMC on breastfeeding status, Hake-Brooks and Anderson (2008:151) reported that KMC mothers breastfed their infants for significantly earlier and longer than the controls (5.08 months vs 2.05 months) ( $P=0.003$ ), and chose to breastfeed more exclusively than the control group (0.04). KMC therefore increases the chances of early initiation of breastfeeding and its continuation for longer periods after discharge from hospital. It also helps

reduce the chances of mixed feeding, thus ensuring that the new born infant gets proper nutrition for adequate growth and development.

### **2.13 SKIN-TO-SKIN CARE AND AND MOTHER- INFANT INTERACTION**

SSC has been shown to have a positive impact on the mothers of preterm infants. KMC provides mothers with the opportunity to know and understand their preterm infants during their stay in the neonatal intensive care unit. They learn to understand their infants sleeping, crying and feeding patterns quicker and better than when the infants are in incubators (Roller, 2005:216).

A qualitative study in which 18 mothers were interviewed after they had spent one hour doing SSC with their preterm in the intensive care unit concluded that maternal confidence developed during the skin-to-skin process was related to their determining of the infant's needs (Johnson, 2007:571). Mothers practicing KMC with their preterm infants have been observed to be more responsive to their infants by touching, gazing at and whispering to their infants during breastfeeding than those whose infants are in incubators (Tessier et al., 1998:5; Tallandini & Scalembra, 2006:265), and have a higher sense of their role as mothers as well as being competent in caring for their infants both in hospital and after discharge (Jefferies, 2012:142). Mothers practicing KMC in hospital have been observed to have significantly lower levels of stress at the time of discharge from hospital than mothers' whose infants were in incubators ( $P < 0.001$ ) (Tallandini & Scalembra, 2006:265).

The interaction between a mother and her preterm infant after hospital discharge has been shown to benefit greatly from the practice of SSC while in hospital. When their infants were three months of age, parents of infants nursed in KMC were found to be more conscious and aware of their child's needs compared to those whose infants were nursed in incubators ( $P < 0.01$ ) (Feldman et al., 2002:20). At six months it was observed that infants who were in the KMC group ( $n=29$ ) spent more time displaying positive behaviour, such as smiling, laughing and reaching out to their parents (28.5% of the 2-minute observation time), compared to infants in the blanket group ( $n=24$ ) (9.6% of the 2-minute observation time) (Neu & Robinson, 2010:407).

Home visits by a nurse when infants were one year old to observe mother-infant interaction noted that SSC within the first 24 hours of the infants life correlated significantly with maternal sensitivity towards infants needs ( $P=0.01$ ) (Bigelow et al., 2010:366). Although mothers have reported that continuous KMC is tiring especially at night, no mother wished to discontinue KMC before the appropriate time (Blomqvist & Nyqvist., 2011:1472).

## **2.14 PATERNAL CARE AND BONDING**

Fathers' presence during childbirth and early contact with the infant immediately after birth is very important in creating and strengthening the father-child relationship later in life (Rodholm, 1981:79; Palkovitz, 1985:299; Vehvilainen-Julkunen & Liukkonen, 1998:10; Grossmann et al., 2002:301). Father's participation in the care of their child is an activity which many men enjoy doing. Fathers whose infants are admitted to the neonatal intensive care unit have reported that the first time they are allowed to hold their infants fills them with joy and a need to protect the infant (Lundqvist, Westas & Hallstrom, 2007:492). Levy-Shiff et al., (1990:289) demonstrated that fathers who visited their preterm infants more frequently in the hospital were more likely to regard these infants in a positive light and to enter into more extensive positive interchange with their infants.

Although fathers are less involved in care taking activities than mothers, they are sensitive to infant cues and are as nurturing as mothers during the interactions with their infants (Parke & Sawin, 1976:369). They have been shown to have the desire to participate in many activities in their infant's' lives, such as visiting the clinic, which gave them the opportunity to gain more knowledge about their children, and they report feelings of pleasure, love and being connected while interacting with and caring for their infants (Garfield & Isacco, 2006:637).

Although fathers regarded their presence in the child's life to be as important as that of the mother, they view their presence to have a completely different impact on the child (Wilson & Prior, 2010:1403). Taubenheim (2006:261) reported that fathers who engaged in caretaking activities such as infant feeding had the most bonding behaviours, suggesting that such activities may be

important elements in the process of paternal-infant bonding. Long hospital stay by mothers has been singled out as a negative aspect of KMC (Lima et al., 2000:25). This can be alleviated if fathers participate in SSC when the mothers are tired.

## **2.15 SUMMARY**

Hormonal changes have been clearly demonstrated to be involved in the onset and maintenance of mammalian maternal behaviour during pregnancy, at birth and thereafter. Little is known about the hormonal changes men undergo as expectant fathers before and after the birth of their infants and how these hormonal changes may affect their parenting behaviour. Few studies have been carried out on human paternal behaviour; most of the knowledge on hormonal involvement in parenting and paternal care behaviours is from animal studies.

There are controversies regarding the involvement of both prolactin and testosterone in male parenting. Evidence relating to the role of these hormones in the promotion of paternal care is conflicting. To date, there has been no published study in which the fathers have had SSC with their infant immediately after birth, with monitoring of the effect of this close contact on hormone changes.

This study is designed to determine the changes in the levels of prolactin and testosterone in first time fathers when they have SSC with their babies within the first six hours after birth. The next chapter describes the methodology used to conduct the study.

# CHAPTER THREE: Research methodology

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## 3 INTRODUCTION

This chapter describes the methodology used to conduct the study and includes the research design, setting, population and sampling method. It also includes the ethical principles taken into consideration during the study as well as the limitations of the study.

### 3.1 RESEARCH DESIGN

The aim of the study was to determine the changes in the levels of serum prolactin and testosterone in first-time fathers over time as SSC takes place between them and their new born infant. In order to achieve this aim a quasi-experimental design with one group pre-test and post-test was chosen. This required that two blood samples were collected from the fathers before the SSC was commenced and then four further blood samples were collected during the SSC at 15 minute intervals for a period of an hour.

Prolactin and testosterone are also found in urine and in saliva; however, the concentration of these hormones in urine and saliva have been found to be very low, and the urine and saliva would need to be of high concentration levels before assay (Ziegler et al.,1996, cited in Schradin & Anzenberger, 2004:671). In the study by Schradin and Anzenberger (2004:673) biological validation of urinary prolactin was done by comparing it with plasma prolactin and plasma prolactin levels were found to be 13 times higher than urinary prolactin levels. For these reasons blood samples were used in preference to urine or saliva.

## **3.2 STUDY SETTING**

The study was conducted at the maternity department of a tertiary hospital in Cape Town. This hospital was selected for the study as it is a tertiary-level teaching hospital where many C/S are performed. The average monthly number of C/S performed is 200, with approximately 50% being emergency C/S. Data collection (i.e. collection of blood samples) was conducted in the recovery area where post-caesarean section patients are stabilised before transfer to the postnatal ward. This area has curtains which can be drawn around the patient's bed for privacy, and collection of blood samples took place here with curtains closed around the participant.

### **3.2.1 GAINING ACCESS**

Permission to conduct the study was obtained from the University of Cape Town Faculty of Health Sciences Human Research Ethics Committee (Appendix 1), and subsequently from the hospital medical superintendent and the Department of Obstetrics and Gynaecology of the hospital (Appendix 4). Once permission was granted, the signed letter of approval together with the protocol was presented to the nursing manager in charge of the maternity unit, who issued a signed letter granting permission. The letter together with the protocol was presented to the nurse in charge of the labour ward and operating theatre for access into the labour ward and theatre.

## **3.3 RESEARCH POPULATION**

Polit and Beck (2006:56) define a population as "all individuals or objects with common defining characteristics". The population for the study was first time fathers whose partners had been delivered by C/S. These fathers were chosen with no restriction as to their age, race, and educational level.

First-time fathers were chosen for the study because they do not have any experience of paternal care. As seen from the literature, males who have been fathers before have raised plasma prolactin levels before and after the birth of their infant (Ziegler & Snowdon, 2000:159). First-time fathers of infants born by C/S were selected for the study because the mother of the baby might be

unable to hold and care for the infant during the first few hours after surgery, as she may be drowsy or asleep from the sedation. This gave the fathers an opportunity to hold their infant during the first few hours after the operation.

### **3.3.1 INCLUSION CRITERIA**

- First-time fathers whose infants were delivered at the tertiary maternity centre
- First-time fathers whose infants were born by C/S

### **3.3.2 EXCLUSION CRITERIA**

- First-time fathers of infants who were born by C/S and were admitted to the neonatal intensive care unit immediately post-delivery

## **3.4 SAMPLING METHOD AND SAMPLE SIZE**

Sampling is “the process of selecting a portion of the population to represent the entire population and a sample is a subset of the population” (Polit & Beck, 2006:260). A convenience sampling method was used to select the research participants. A sample size of 15 men was chosen for the study, this sample being chosen because previous studies had used a sample size of 15 participants in determining prolactin levels in males. Ziegler et al. (2009:437) used a sample size of 15 to determine the role of prolactin in parenting. Schradin and Anzenberger (2004:672) used a sample size of 15 to determine prolactin levels in male marmosets becoming first-time fathers. In this study, however, only 11 men were successfully recruited to collect a total of six blood samples from each for the data analysis.

### **3.4.1 STATISTICAL POWER CALCULATION**

The statistical power for the study was initially calculated for a sample size of 15, using the means and SD values of a study conducted by Ziegler et al. (2009) to determine changes in prolactin levels over time with a sample size of 15. However, only 11 fathers were successfully incorporated into the study. The statistical power for a sample size of 11 is as follows;

.sampsi 9.3 5.6, sd1 (2.4) sd2 (1.0) n1 (11) n2 (11)

Estimated power for two-sample comparison of means

Test Ho:  $m_1 = m_2$ , where  $m_1$  is the mean in population 1 and  $m_2$  is the mean in population 2

Assumptions:

Alpha= 0.0500 (two-sided)

$m_1 = 9.3$

$m_2 = 5.6$

SD1= 2.4

SD2= 1.0

Sample size  $n_1 = 11$  and  $n_2 = 11$

Estimated power: 0.9997

Where:

Mean value and SD of prolactin before start of SSC is 9.3 and 2.4

Mean value and SD of prolactin at 60 minutes of SSC is 5.7 and 1.0

### **3.5 RECRUITMENT OF PARTICIPANTS**

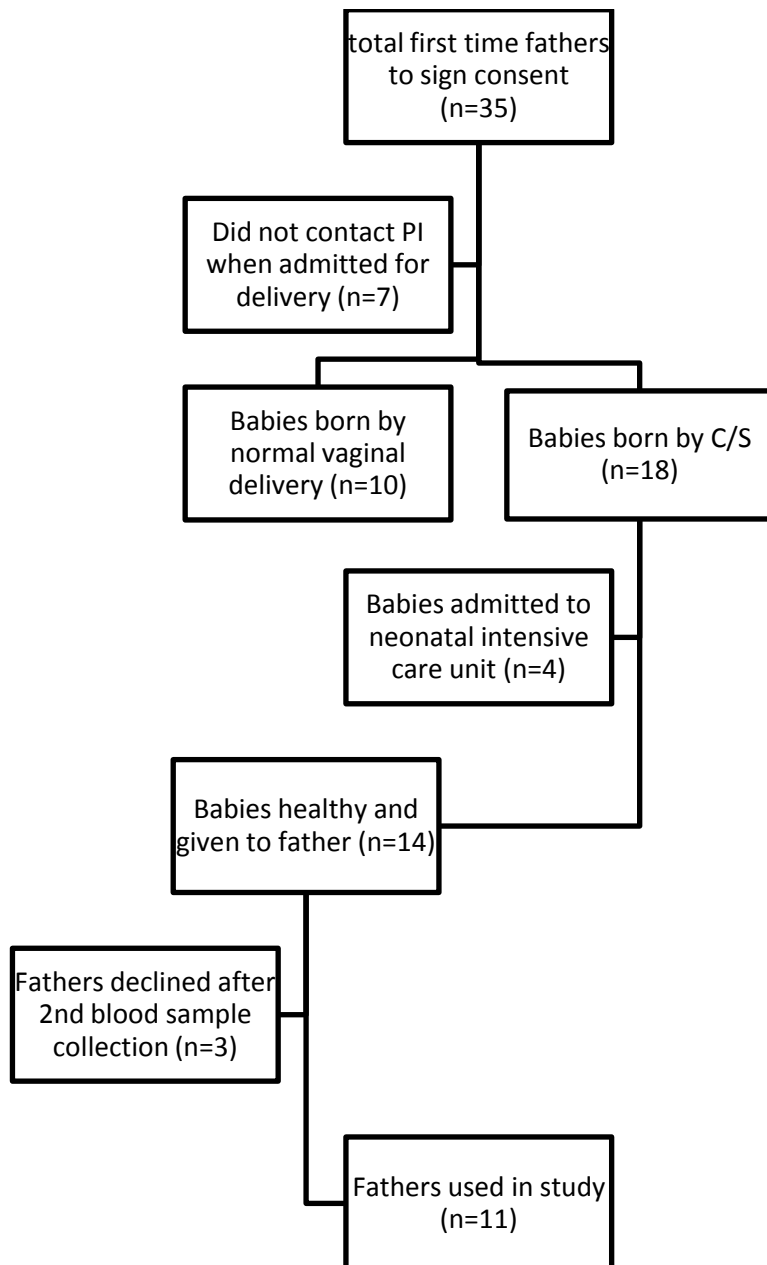
After permission was granted to conduct the study and access to the hospital's maternity section was granted, the researcher proceeded to inform women attending the ante-natal clinics at the maternity centre about the study. Every morning, before talking to the women at the clinic, the study was explained to the staff at the clinic and the letter granting permission to conduct the study in that setting was presented to them.

While the women were waiting in the lounge area, the researcher explained the study. The study was first explained in English, and then in Afrikaans and isiXhosa (the other official languages in the Western Cape Province). The women were informed that participation in the study was voluntary and that there would be no compensation to them or their partners for taking part in the study. They were then given opportunities to ask questions about the study. An information sheet (Appendix 2) about the study in the language of choice was provided. This described the purpose of the study, potential risks/benefits, the right to confidentiality and the right to withdraw at any time.

The women were requested to take this home and discuss the study with their partners. They were asked to contact the researcher if they and their partners were interested in taking part in the study.

When the couples who wished to take part in the study made contact with the researcher, an arrangement was made to meet them to discuss any questions and provide further information. Couples who agreed to participate were asked to sign a consent form (Appendix 3) of which they were given a copy. They were asked to contact the researcher on the day that the woman was to give birth. The researcher then met the couple at the hospital.

The process of informing the women at the clinic about the study took place on Tuesdays and Thursdays, the clinic days at the maternity centre, from February to August 2011. A total of 35 first-time fathers agreed to participate and signed consent. Of this number, seven fathers did not contact the researcher when their partner was admitted for delivery. The infants of ten fathers who contacted the researcher were born by normal vaginal birth. The infants of four fathers who were born by C/S were admitted to the neonatal intensive care unit and three fathers withdrew during the data collection process (Figure 1). Although most of the pregnant women had indicated that they would want their partners to participate, some of the men were reluctant to have blood samples taken.



**FIGURE 1: Study flow chart to obtain the 11 participating first-time fathers**

The figure above is a diagrammatic representation of the steps involved in obtaining a sample size of 11 fathers in this study.

### **3.6 DATA COLLECTION**

The researcher met with the couples at the hospital and they were again reminded what the study was about, the risks and benefits, and their right to withdraw at any time. Once the potential participants had agreed to continue, the researcher informed the primary care-givers of the women (doctors and/or midwives) about the study, to confirm that the study would not in any way interfere with the care of the woman as well as the care of the child to be born.

While the woman was being prepared for the C/S, the partner was taken to theatre and given theatre clothes to wear. He was then taken to the recovery area in the theatre and seated comfortably in an arm-chair. He was reminded that he could withdraw at any moment if he felt uncomfortable. Curtains were drawn around him for privacy. An intravenous catheter was inserted into the peripheral vein of his arm and the first blood sample was taken. He was then taken into theatre where he sat beside his partner while the C/S was being performed.

The second blood sample was taken just as the infant was born. After the paediatrician had assessed the infant and confirmed that the infant could stay with its father, the father was taken back into the recovery area and the infant was put on his chest. The infant wore only a nappy to prevent soiling the father with urine or stool. The father wore no shirt or vest to ensure that he and the infant were in SSC. A special shirt together with an inner cloth (kanga carrier) was used to secure the infant on the father's chest.

The third blood sample was taken 15 minutes after SSC commenced. The fourth, fifth and sixth blood samples were collected every 15 minutes thereafter up to the period of one hour. Each time 5 ml of blood was collected, making a total of 30 ml of blood collected from each participant. A data collection sheet (Appendix 5) was used to enter the details of the participants and the time and number of blood samples taken.

### 3.7 DATA MANAGEMENT AND ANALYSIS

The blood samples were collected in test tubes with vacuum seal containing Serum Sep activator (yellow cap) and inverted five times to mix the blood. The test tubes were labelled with date, time and the study number of the participant. The samples were then left at room temperature for about five minutes to clot. The blood samples were then kept in a cooler box on ice until they were transported to the laboratory, where they were spun in a centrifuge with the temperature controlled at 4°C at 3000 rpm for 10 minutes. The serum was then pipetted into safe-lock tubes labelled with date, time and study number of participant, and stored at -80°C in an upright position until analysed in an appropriately equipped laboratory.

At the laboratory a quality control test was done using the reagents provided specifically to monitor the system performance and chart trend. The specimens were defrosted and checked to make sure the serum was free of air bubbles and any fibrin or particulate matter. They were then placed into the Advia Centaur system together with the reagents for analysis. When the reaction was completed, the system reported the results. These results were entered into Excel spreadsheets by the laboratory technician, and cross-checked by the researcher. These values were then transferred into the Statistica statistical and data analysis program by the researcher for further analysis.

Given that the sample size in this study was small ( $n=11$ ), with 66 observations each for prolactin and testosterone, it was assumed that the variables would not be normally distributed; therefore non-parametric methods of statistical analysis were used in the analysis of the data (Corder & Foreman, 2009:2). The median, maximum value, minimum value and interquartile range, together with the means and SD were used for descriptive statistics. The Wilcoxon matched pairs test, which is the alternative to the  $t$ -test in parametric statistics for dependent variables, was used to compare the variables and to test for probability (Kimble et al., 2004:210; Corder & Foreman, 2009:2).

### **3.8 VARIABLES**

The characteristics, qualities, or properties of persons, things, or situations that are measured and about which data are collected and that change or vary are known as variables (Burns & Grove, 2001). The variables used in this study are discrete numerical variables which are prolactin values and testosterone values.

#### **3.8.1 DEPENDENT VARIABLES**

There are two dependent variables:

- The level of serum prolactin in fathers before and during SSC;
- The level of serum testosterone in fathers before and during SSC.

These variables are the dependent variables because they are expected to be affected by the SSC.

The levels of serum prolactin and testosterone are expected to change during the SSC process.

The extraneous variable of the study is time. The blood samples were collected at different times of the day since the infants were born at different times.

### **3.9 PILOT STUDY**

This study was a parallel study modelled on one that was being conducted by Dr Nils Bergman, who had tested the practicalities and the routines in an ongoing study (REC Ref. 350/2008). He was also present to assist with the first case. Therefore a pilot study was not conducted.

### **3.10 MEASUREMENT INSTRUMENTS**

The Siemens Medical Solutions Diagnostics instrument with the trademark 'ADVIA Centaur' was used for measurement in this study. Prolactin and testosterone assays were conducted using this instrument.

### **3.10.1 RELIABILITY OF INSTRUMENT**

The ADVIA Centaur prolactin assay standardisation is traceable to WHO third IRP (international reference preparation) for human prolactin (84/500). A comparison of the full assay range gave the following correlation: ADVIA Centaur prolactin=1.06 (WHO) -0.6IU/ml,  $r=0.99$  (Siemens Medical Solutions Diagnostics, 2007).

The ADVIA Centaur testosterone assay is standardised using internal standards manufactured analytically which are traceable to gas chromatography-mass spectroscopy (GCMS). The following equation describes the relationship between testosterone standards and GCMS analysis throughout the range of the assay: ADVIA Centaur Testosterone=1.00 (GCMS) +1.35 ng/dl,  $r=1.00$  (Siemens Medical Solutions Diagnostics, 2007).

To ensure that the same quantity of blood was collected, only 5 ml syringes were used to draw blood from the participants. The blood was slowly pipetted into the test tubes while slanting the test tubes to prevent the blood from haemolysing and preventing the red blood cells from contaminating the serum after the blood was centrifuged. The serums were kept in an upright position in the deep freezer to prevent the formation of bubbles.

### **3.10.2 VALIDITY OF INSTRUMENT**

The ISO/IEC 1705 are the general requirements for the competence of testing and calibration laboratories, which were used in the laboratory by the technician to ensure validity of the instrument. Two levels of this material were assayed each day before the blood samples were analysed. The ISO/IEC 1705 has an insert with the suggested expected values, if the quality control values do not fall within the expected values, the following actions are taken:

- It is verified that the material has not expired;
- The required maintenance is performed;
- It is verify that the assay was performed according to instructions; and
- The assay is rerun with fresh quality control products.

### **3.11 ETHICAL CONSIDERATIONS**

The following ethical principles adapted from the Declaration of Helsinki developed by the World Medical Association (2013) were observed in order to adhere to research ethical considerations.

#### **3.11.1 AUTONOMY**

Respect for autonomy means participants must make free and informed choices to participate in a study (Polit & Beck, 2006). The researcher provided the participants with an information sheet which explained the purpose of the study, the study method, risks/benefits, right to withdraw and data collection procedure. Repeated explanations of this information were provided to the participants throughout the study. They were given time to reflect on the study and to discuss with their partners and family before consenting to participate.

#### **3.11.2 BENEFICENCE**

The researcher explained to the participants that there was no direct benefit to them; however, the process of SSC gave them an opportunity to bond with their infant. The process also helped to stabilise their infant, which is reported to have a positive impact on the infant's physiological development. They were also informed that there were to be no incentives for taking part in the study. However, the 'kanga-carrier' was given to them after the data collection was completed to enable them to continue SSC at home.

#### **3.11.3 NON-MALEFICENCE**

The risk of infection was minimised through the use of sterile materials during the process of blood collection. Aseptic techniques were used during the collection of blood samples, such as cleaning the area with alcohol swabs before inserting the needle, and placing a clean dry dressing on the area after removing the intravenous catheter when blood collection was completed. Discomfort felt during insertion of the intravenous catheter was minimised by inserting it only once and then using a three-way stop-cock which was opened and closed each time a blood sample was collected. The fathers were seated comfortably in an arm chair during the process of SSC and collection of blood.

#### **3.11.4 JUSTICE**

The principle of justice includes the participant's right to fair treatment and privacy (Polit & Beck, 2006). There was no discrimination during the selection of the participants, couples who were willing to be part of the study and met the inclusion criteria, regardless of race, economic status, age and educational level, were included in the study. All participants were treated fairly and equally before, during and after the study. Five of the participants requested to know their results and these were delivered to them in person by the researcher.

#### **3.11.5 RIGHT TO PRIVACY AND CONFIDENTIALITY**

Confidentiality is a pledge made by the researcher to participants that no information will be made accessible to any third party who is not involved in the study. When no one is able to link the data collected to any of the participants because they are nameless, anonymity is respected (Polit & Beck, 2006). The names of the participants and their partners were known only to the researcher. Coded numbers and letters were used to link the blood samples to the individual participants. To ensure privacy of the participants during the SSC process and collection of blood samples, they were placed in the recovery area of the theatre and curtains were drawn around them. Information obtained from the fathers during the study was not made public to any third party not involved in the study without the permission of the fathers.

### **3.12 LIMITATIONS OF THE STUDY**

A small sample size was used for the study (11 participants) in a single hospital setting, and therefore the results of the study may not be generalised to the broader population.

A convenience sampling method was used to select participants which did not give each subject of the general population an equal chance of being selected.

The extraneous variable of the study, which is time, was not controlled. The blood samples were collected at different times of the day, since the babies were born at different times of the day. The level of blood hormones could vary among the fathers according to the time of the day.

### **3.13 SUMMARY**

A quasi-experimental design with one group pre-test and post-test was used for the study. Ethical approval was obtained and ethical principles were adhered to. Recruitment of study participants and data collection were carried out at the maternity centre after permission to conduct the study was granted. Eleven first-time fathers participated in the study.

# CHAPTER FOUR: Presentation of results

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## 4 INTRODUCTION

This chapter presents the data collected and shows how they were analysed to obtain results. The values of prolactin and testosterone obtained from the assay were analysed using the STATISTICA statistical and data analysis program. The aim of this study was to determine changes in the levels of serum prolactin and testosterone when first-time fathers had SSC (kangaroo care) with their new born infants after birth by C/S. The analysis of prolactin was done separately from that of testosterone; each hormone was analysed for changes in its value as SSC continued.

### 4.1 DEMOGRAPHIC DATA

Eleven fathers volunteered to participate in the study and six blood samples were collected from each participant. The relationship status between the parents of the new born infant, the age of the father and the type of C/S performed were included in the demographic data. The blood samples collected from these fathers were analysed using prolactin and testosterone assays, and tables 1 and 2 show the values of prolactin and testosterone obtained after analysis of the six blood samples taken from each father.

#### 4.1.1 TYPE OF C/S

Of these 11 fathers, 45% (n=5) of them had a partner whose infant was delivered by elective C/S and 55% (n=6) had a partner whose infant was delivered by an emergency C/S. An elective C/S is one which is planned in advance and is performed on a set date, while an emergency C/S is performed due to unforeseen problems during labour, for example foetal distress.

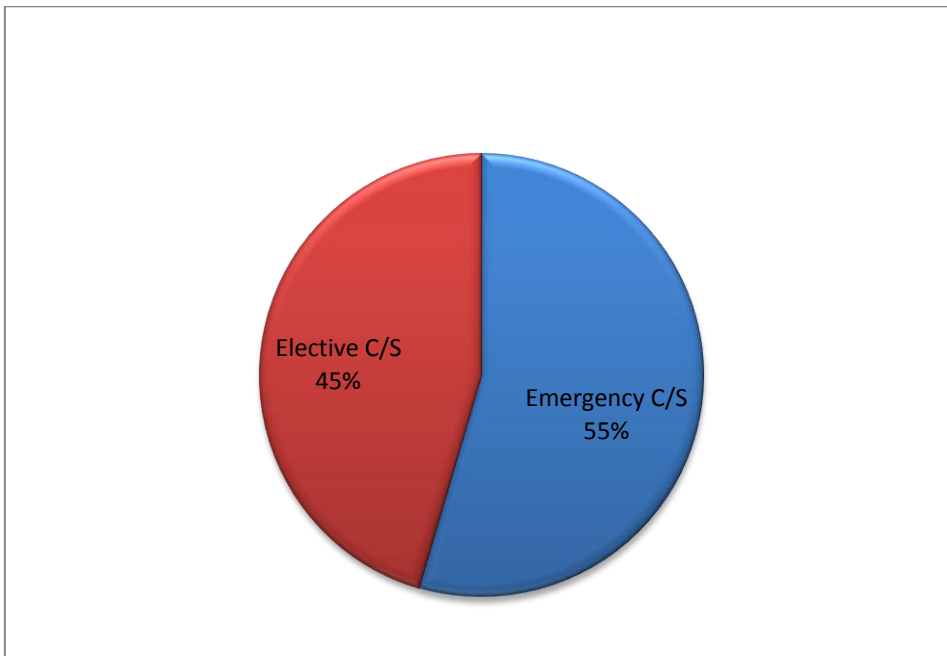


Figure 2: Type of C/S

#### 4.1.2 RELATIONSHIP STATUS

Figure 3 shows the relationship status of the parents: 45.4% (n=5) of the fathers in the study were married to the mother of their child, and 36.4% (n=4) were engaged to the mother while 18.2% (n=2) were in a dating relationship with the mother of their child.

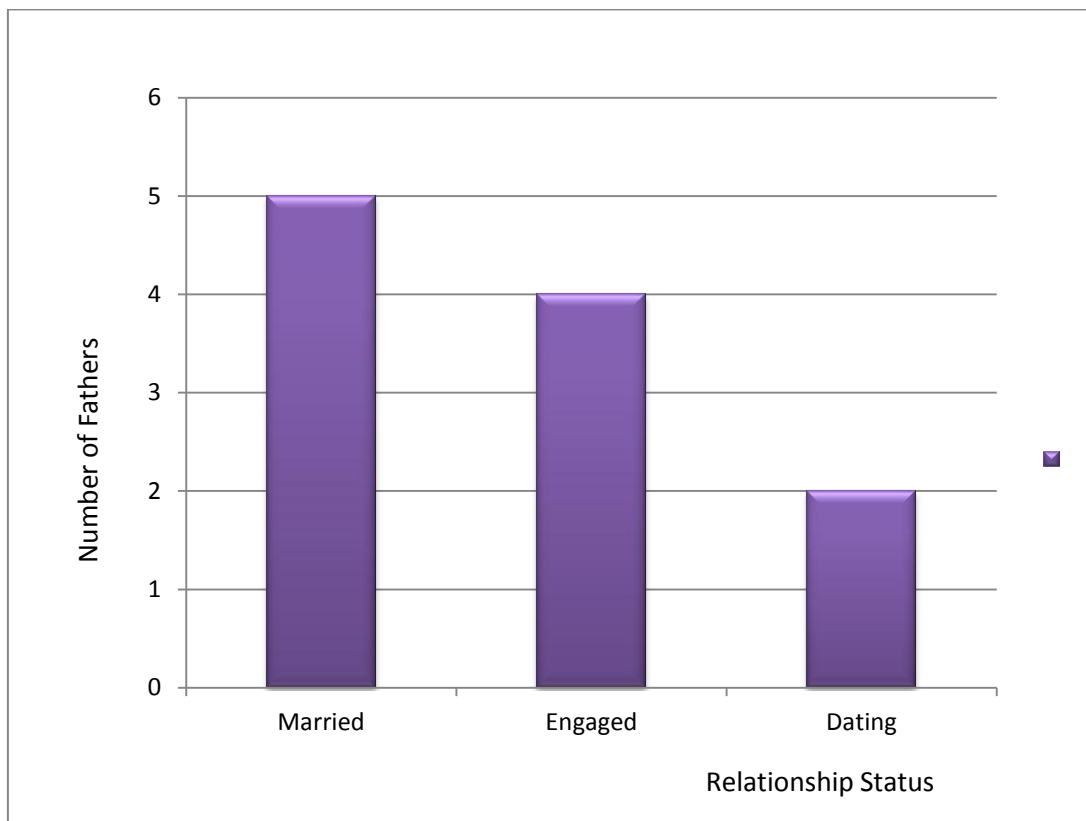


Figure 3: Parent's Relationship Status

### 4.1.3 AGE DISTRIBUTION

The age distribution of the fathers is shown in Figure 3. All of the fathers included in the study were aged between 24 and 32 years: 27% (n=3) of the fathers were aged 28; four (9%) were aged 24, 25, 27 and 29 respectively. Two fathers were aged 30 (18%) and two were aged 32 (18%).

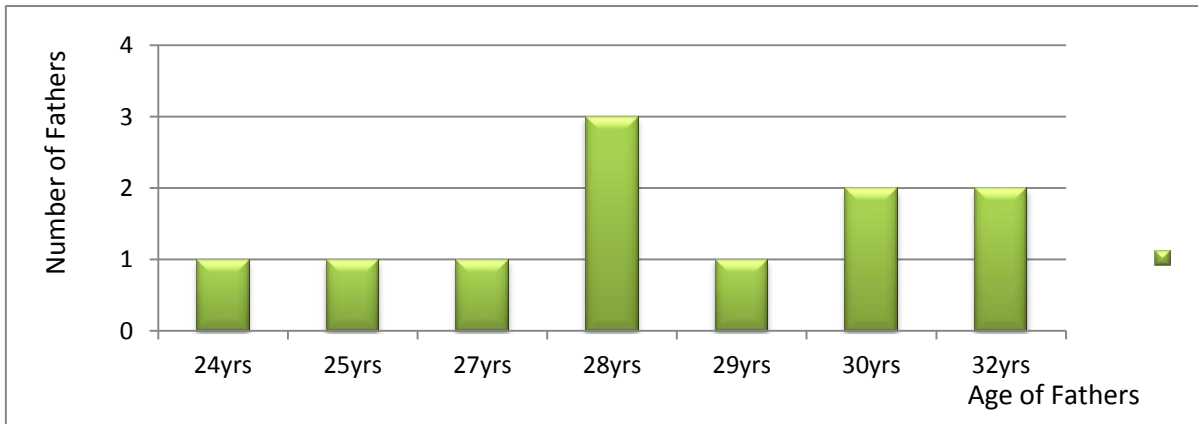


Figure 4: Age of the Fathers

**Table 1: Values of prolactin (ng/l) obtained from analysis of blood samples**

Specimen number	1	2	3	4	5	6
ID	PRL_PRE	PRL_START	PRL_SSC15	PRL_SSC30	PRL_SSC45	PRL_SSC60
P001	8.93	8.34	6.4	6.29	5.5	5.95
P002	6.33	5.43	6.7	5.99	4.96	4.7
P003	8.55	14.02	1151	11.86	10.55	15.38
P004	10.39	12.69	10.02	10.07	8.43	7.6
P005	10.04	12.81	7.92	6.18	4.94	5.74
P006	19.17	21.68	24.55	22.08	20.06	17.44
P007	8.33	7.43	7.39	7.25	7.72	9.32
P008	10.98	13.23	11.26	10.0	8.82	12.11
P009	8.56	7.73	7.57	7.03	6.7	6.08
P010	5.03	6.31	5.72	4.01	4.62	4.93
P011	4.04	5.5	4.42	3.4	4.92	4.19

**Table 2: Values of testosterone (nmol/l) obtained from analysis of blood samples**

Specimen number	1	2	3	4	5	6
ID	TES_PRE	TES_START	TES_SSC15	TES_SSC30	TES_SSC45	TES_SSC60
P001	16.49	16.48	16.99	21.84	20.31	20.84
P002	18.41	10.97	21.47	17.65	17.6	17.72
P003	21.47	22.0	11.51	13.55	14.69	8.56
P004	9.31	12.61	10.39	12.99	12.66	12.72
P005	14.85	16.89	12.06	9.33	13.67	12.05
P006	10.91	9.53	11.24	11.63	12.94	13.44
P007	15.49	16.24	19.23	17.39	19.9	19.64
P008	6.24	7.46	8.09	9.65	11.04	11.36
P009	13.63	19.59	20.4	18.95	16.96	17.23
P010	6.67	6.52	5.96	5.62	6.46	5.63
P011	13.6	14.4	13.93	13.23	14.34	10.73

## 4.2. SUMMARY OF STATISTICS FOR PROLACTIN

Eleven fathers each gave six blood samples (N=66). Figure 5 below shows the distribution of prolactin obtained from all blood samples of the eleven participants. The mean value is 8.9ng/l with a 95% confidence interval between 7.842 and 10.15. The standard error mean value is 0.576, the median value is 7.6ng/l, and the inter-quartile range is 5.7 - 10.5 ng/l, minimum value 3.4ng/l, maximum value 24.4ng/l and Standard Deviation 4.679.

Laboratory reference values of serum prolactin in adult males range between 2.1 ng/l and 17 ng/l with a mean value of 7 ng/l (Siemens Medical Solutions Diagnostics, 2007). Most prolactin values in the study ranged between 5.7 ng/l and 10.5 ng/l which is within the range of the laboratory reference values.

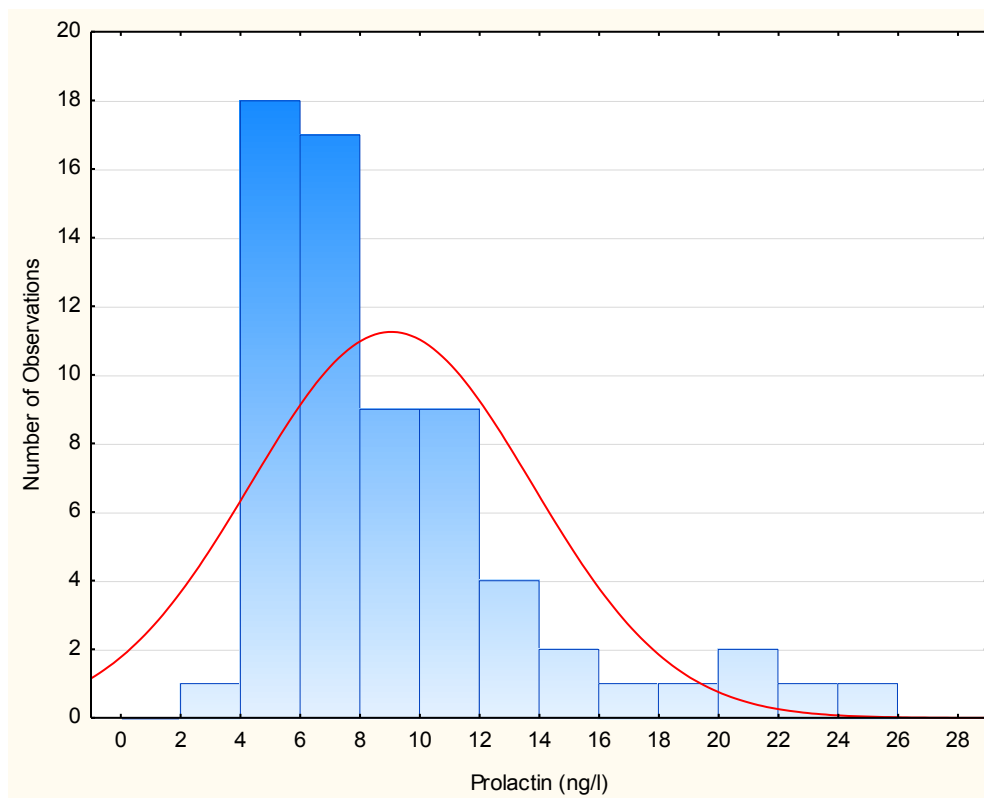


Figure 5: Distribution of prolactin levels

### 4.3 ANALYSIS OF PROLACTIN TRENDS

Figure 6 shows the progressive change in the values of prolactin for each father from the first blood sample collection to 60 minutes of SSC with their infant. Each father shows a different variation in the values of serum prolactin as SSC progresses. One of the fathers (P006) has significantly higher prolactin values compared to the other fathers, his prolactin levels increased to the maximum level after 15 minutes of SSC, after which they dropped progressively to the end of SSC at 60 minutes.

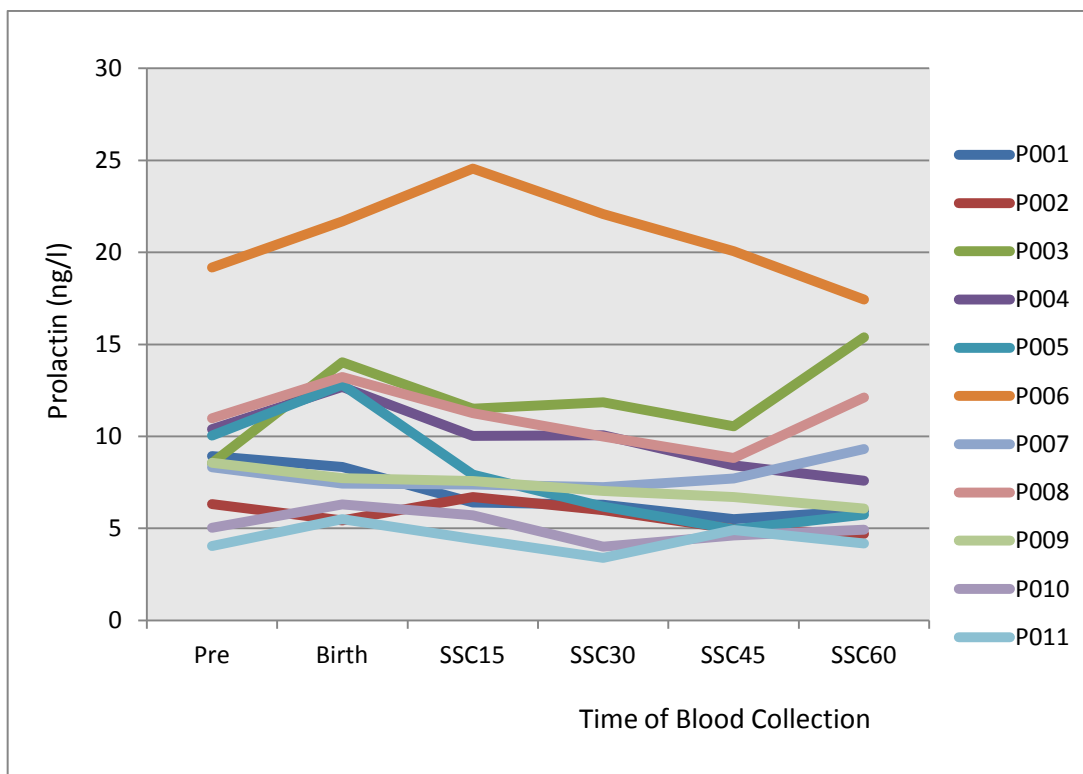


Figure 6: Prolactin Trends for Each Father

prolactin levels grouped by time of blood collection are shown in Figure 7, with median, interquartile range, maximum and minimum values of prolactin at each time of blood collection in the fathers. One of the fathers had very high prolactin levels and is seen as an outlier. In analysing the change in prolactin as SSC progressed, this father's prolactin values were excluded to prevent biasing the results.

Figure 7 shows that the prolactin levels increases at point 2 and then drop progressively to point 5, after which they increases slightly at point 6. Using the Wilcoxon matched pairs test to compare prolactin values from the time of first blood collection (point 1) to the time of delivery of the infant (point 2) gives a P value of 0.04, indicating that the increase in prolactin levels is significant. Comparing the values of prolactin from point 1 to those at 60 minutes of SSC (point 6) gives a P value of 0.2, implying that the decrease in prolactin levels after 60 minutes of SSC is insignificant. The progressive decrease in prolactin from the time of birth (point 2) to 45 minutes of SSC is significant ( $P=0.004$ ). The prolactin levels increase between 45 minutes of SSC and 60 minutes of SSC, but this increase was insignificant ( $P=0.5$ ).

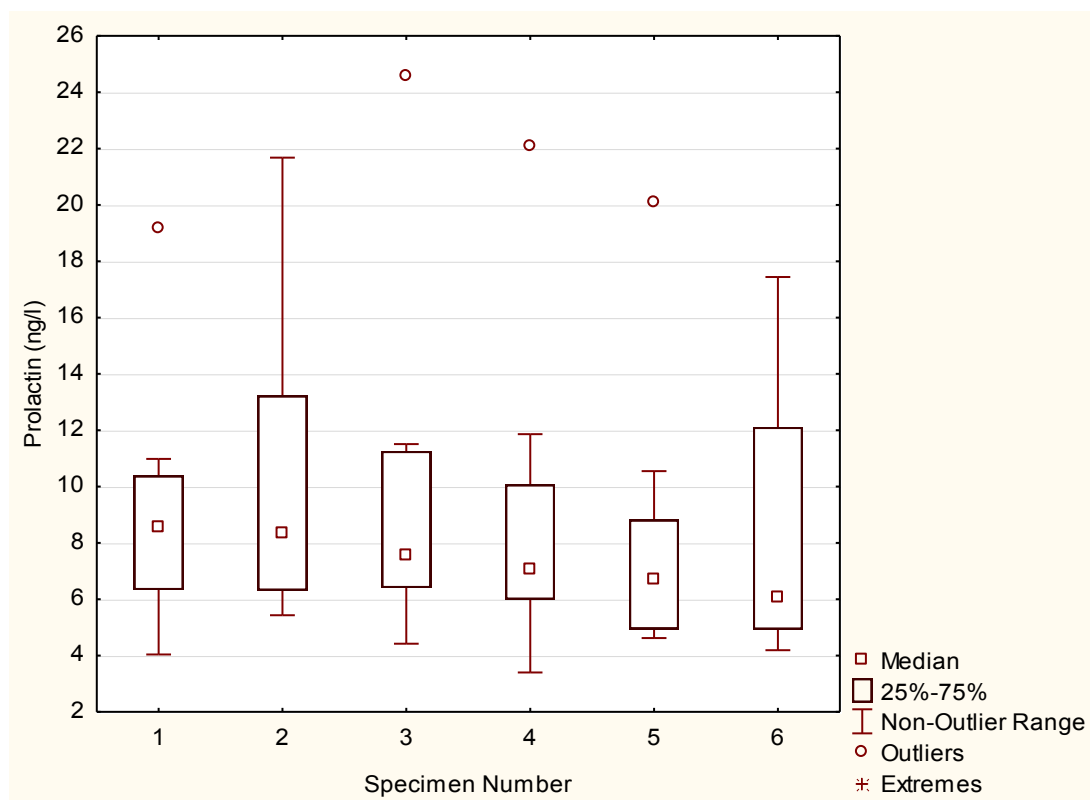


Figure 7: Prolactin levels grouped by time of blood collection

### 4.3.1 PROLACTIN VARIATION AND TYPE OF C/S

The trend of median prolactin values with SSC of the fathers as grouped by type of C/S performed is shown in Figure 8. Fathers of infants born by emergency C/S have higher median prolactin values than fathers whose infants were born by elective C/S. Prolactin levels increase in both groups of fathers at the time of birth, although this increase is insignificant ( $P=0.1$  for emergency C/S and  $P=0.4$  for elective C/S). These values then drop progressively until 45 minutes of SSC ( $P=0.06$  for emergency C/S,  $P=0.5$  for elective C/S), and are both insignificant.

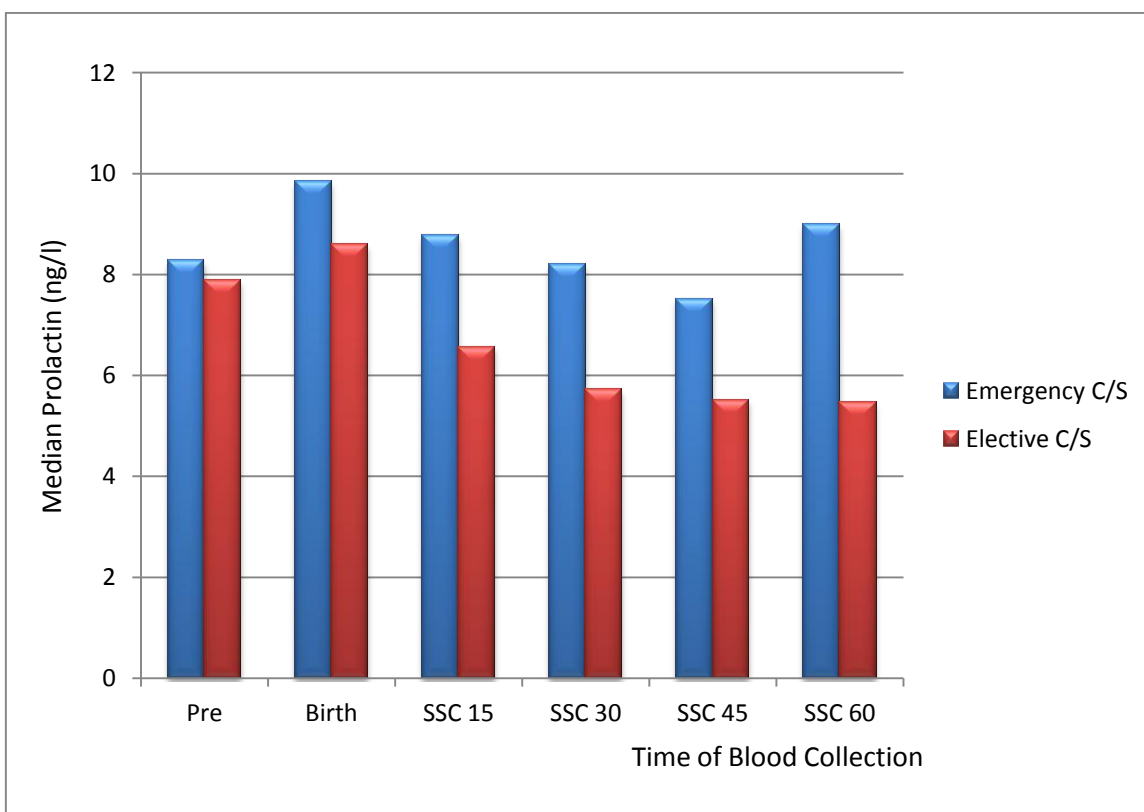


Figure 8: Prolactin trend of fathers grouped by type of C/S performed

### 4.3.2 PROLACTIN VARIATION AND RELATIONSHIP STATUS

Figure 9 below shows the progressive change in median prolactin value with SSC of the fathers grouped by their relationship status. Men who are married to the mother of their baby have higher median prolactin values than those who are engaged to or dating the mother of the baby. Men in a dating relationship with the mother of their baby have the lowest median prolactin levels throughout the process of SSC.

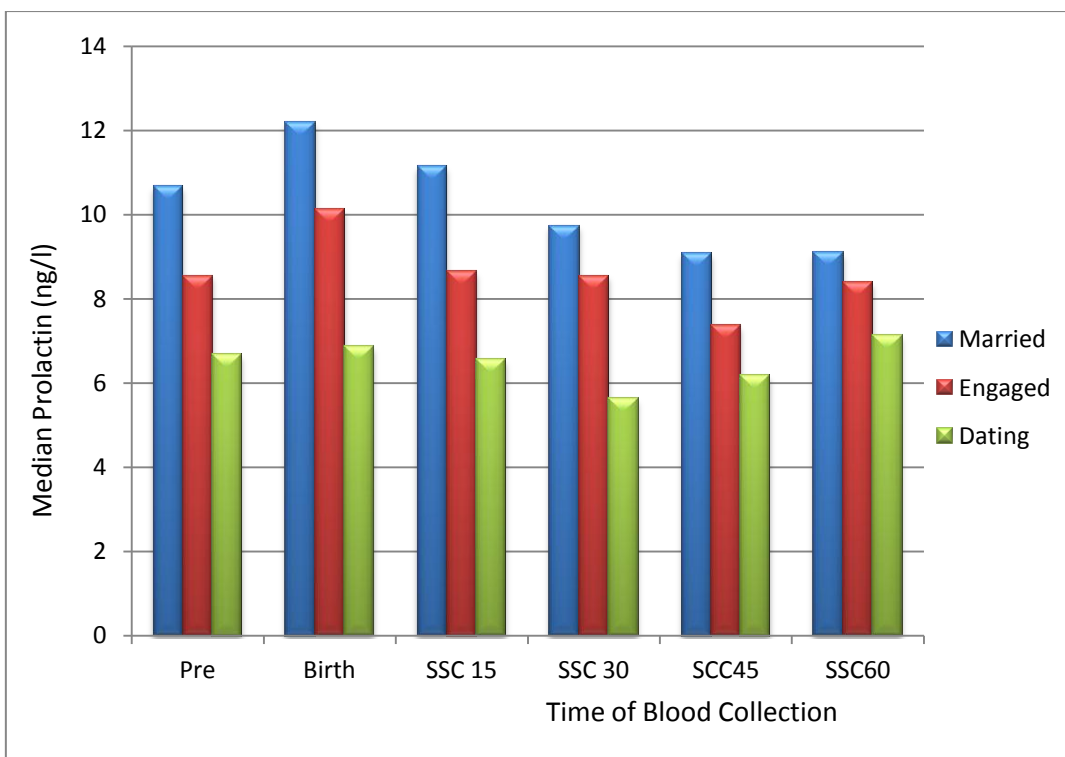


Figure 9: Prolactin trend of fathers grouped by relationship status

### 4.3.3 PROLACTIN VARIATION AND AGE

Median values of prolactin with SSC for fathers grouped according to age are shown in Figure 10. The fathers were divided into approximately two equal groups since it is median values that are used: 28 years and below (six fathers) and 29 years and above (five fathers). Median prolactin levels in fathers aged 29 years and above are generally higher compared to those of fathers aged 28 years and below. In both groups of fathers the median prolactin level increased at the time of the infant's birth ( $P=0.2$  for fathers aged 28 and below;  $P=0.3$  for fathers aged 29 and above). It then decreased progressively up to 45 minutes of SSC, after which it increased at 60 minutes of SSC ( $P=0.6$  for fathers aged 28 and below;  $P=0.2$  for fathers aged 29 and above).

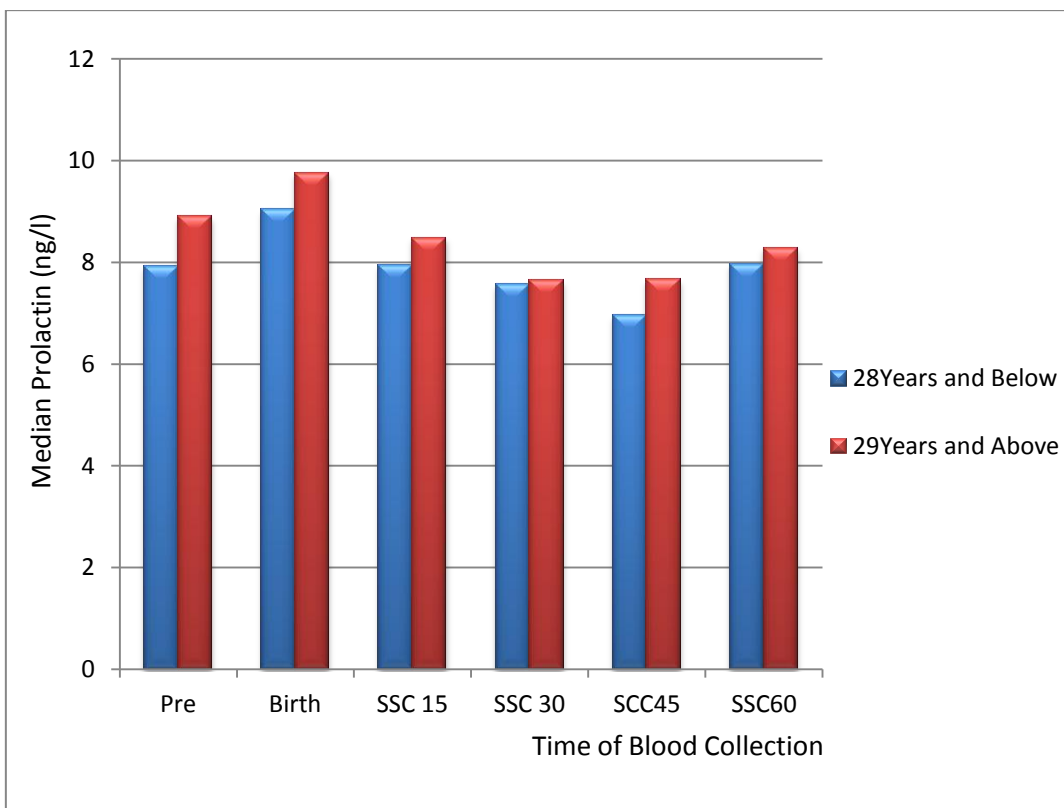


Figure 10: Prolactin Trend of Fathers Grouped By Age

#### 4.4 SUMMARY OF STATISTICS FOR TESTOSTERONE

The distribution of testosterone levels obtained from all six blood samples from all the eleven participants is shown in Figure 11. The mean value is 13.8 nmol/l, with a standard error mean of 0.562 and Standard Deviation of 4.566, with a skew value of -0.0174. The median value is 13.5 nmol/l, with an interquartile range of 10.9 - 17.3 nmol/l, a minimum value of 5.6 nmol/l, maximum value of 22.0 nmol/l, and 95% confidence interval of 12.75 - 14.96.

Laboratory reference values for serum testosterone in adult males ranges between 8.4 nmol/l and 28.7 nmol/l, with a mean value of 15.8 nmol/l (Siemens Medical Solutions Diagnostics, 2007). Most testosterone values in the study range between 10.9 and 17.3 nmol/l, and are within the range of the laboratory reference values.

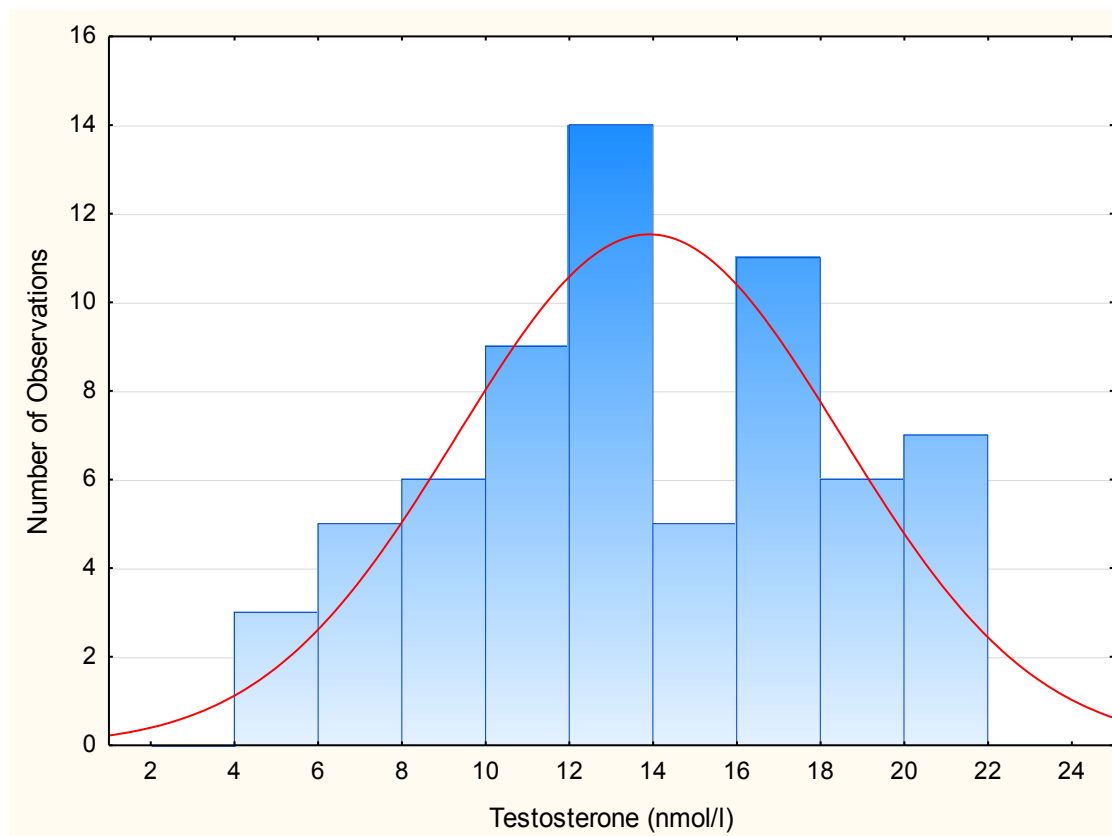


Figure 11: Distribution of testosterone levels

#### 4.5 ANALYSIS OF TESTOSTERONE TRENDS

Figure 12 shows the values of testosterone for each father and how they vary with time. It can be observed that there is a variation of testosterone in each of the fathers at each point of blood collection. Participant P002 shows a sharp drop in testosterone at the time of the birth of his baby, with a sharp increase 15 minutes into SSC followed by decrease at 30 minutes of SSC, and remains stable until 60 minutes of SSC.

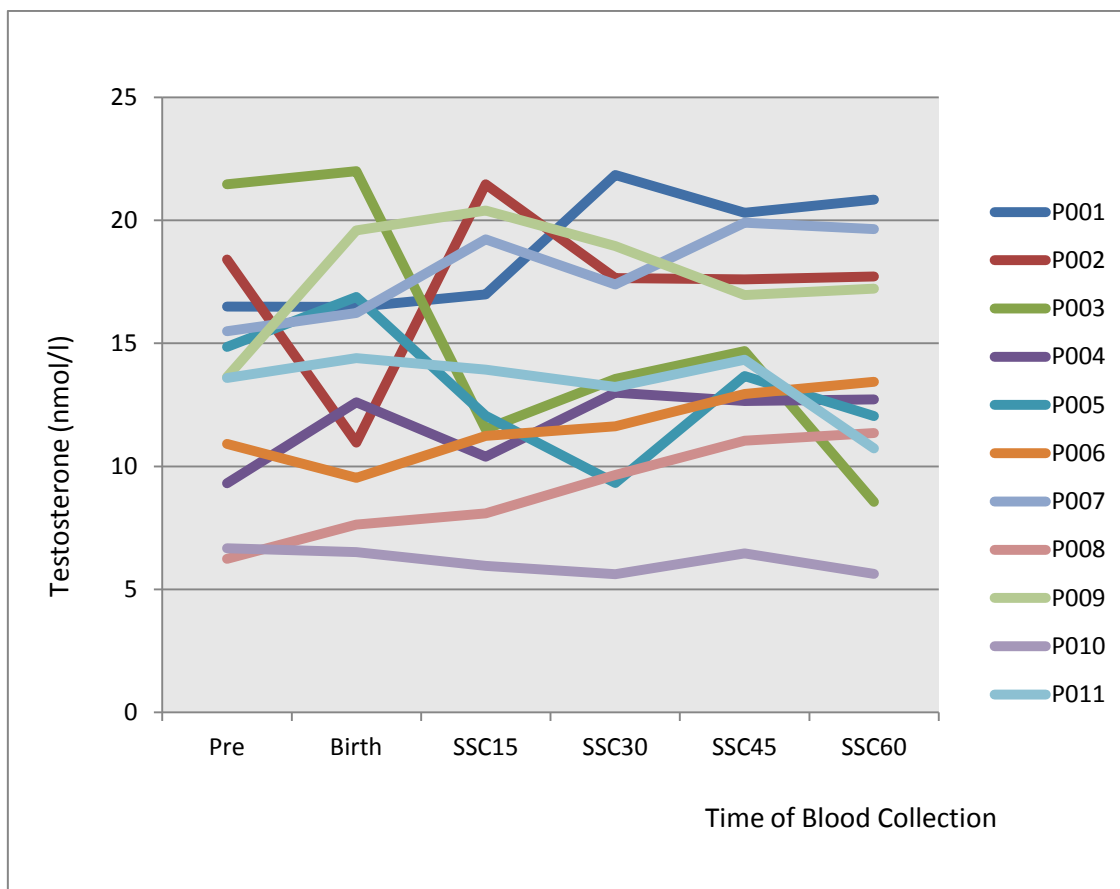


Figure12: Testosterone Trends for Each Father

In Figure 13 testosterone levels are grouped by time of blood collection, showing the median, interquartile range, and maximum and minimum values. Testosterone values show little change throughout, increasing very slightly from point 1 to 2. The Wilcoxon matched pairs test to compare testosterone levels between the time of first blood collection and the time of the birth of the baby shows that this increase was statistically insignificant ( $P=0.2$ ). Testosterone levels increased further after 15 minutes of SSC (point 3), which was also insignificant ( $P=0.3$ ). Its value decreases at 30 minutes of SSC (point 4) ( $P=0.9$ ), after which it remains almost constant at points 5 and 6.

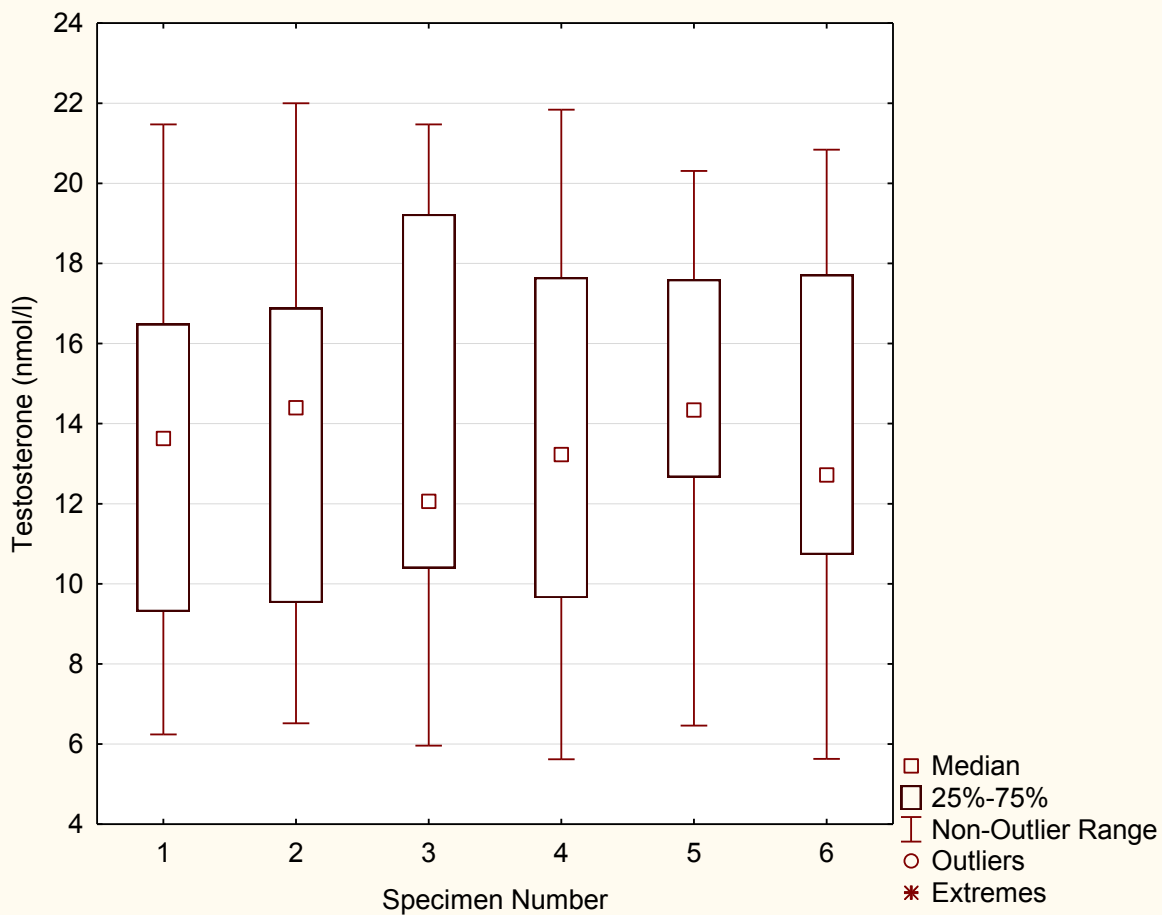


Figure 13: Testosterone levels grouped by time of blood collection

#### 4.5.1 TESTOSTERONE VARIATION AND TYPE OF C/S

The changes in median testosterone levels with SSC in fathers whose infants were born by either an elective C/S or an emergency C/S are shown in Figure 14. Fathers of infants born by elective C/S have higher levels of testosterone throughout the SSC than fathers whose infants are born by emergency C/S. In both groups of fathers the level of testosterone remains almost constant, with minimal changes.

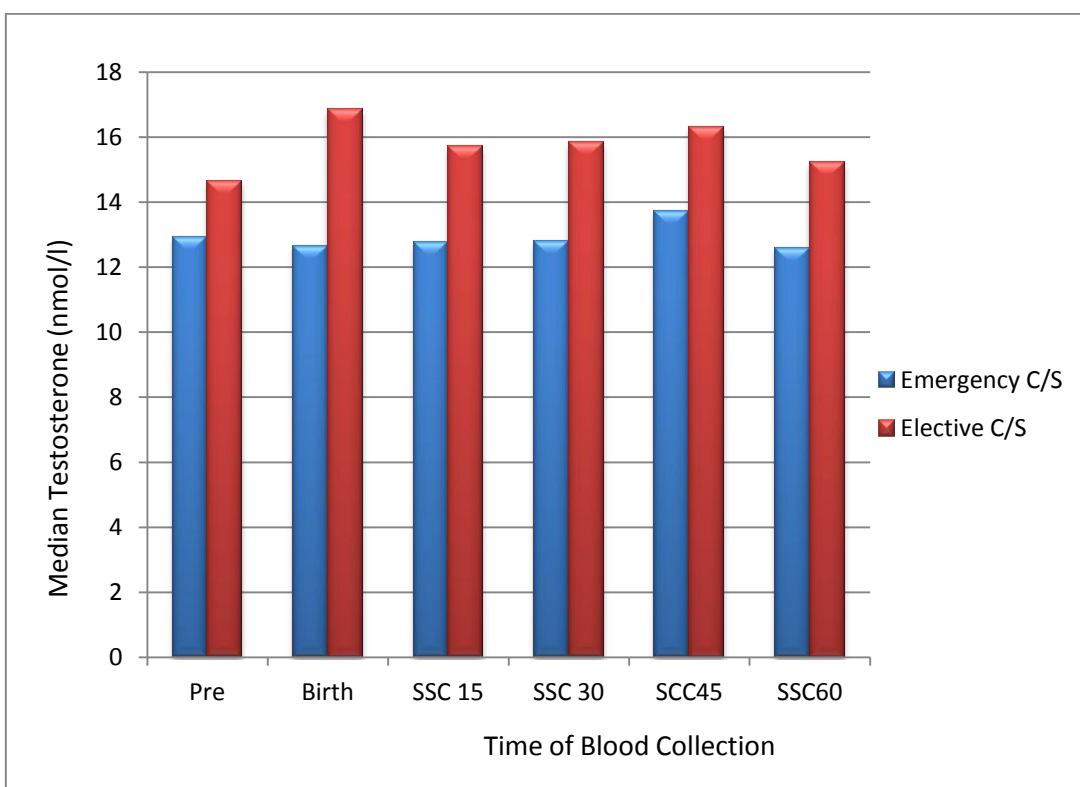


Figure 14: Testosterone trend of fathers grouped by type of C/S

#### 4.5.2 TESTOSTERONE VARIATION AND RELATIONSHIP STATUS

Figure 15 shows the change in the value of mean testosterone with SSC in fathers who were either married or engaged to or dating the mother of their infant. Fathers who were engaged to be married to the mother of their infant had the highest values of mean testosterone from the time of first blood collection right through to 60 minutes of SSC.

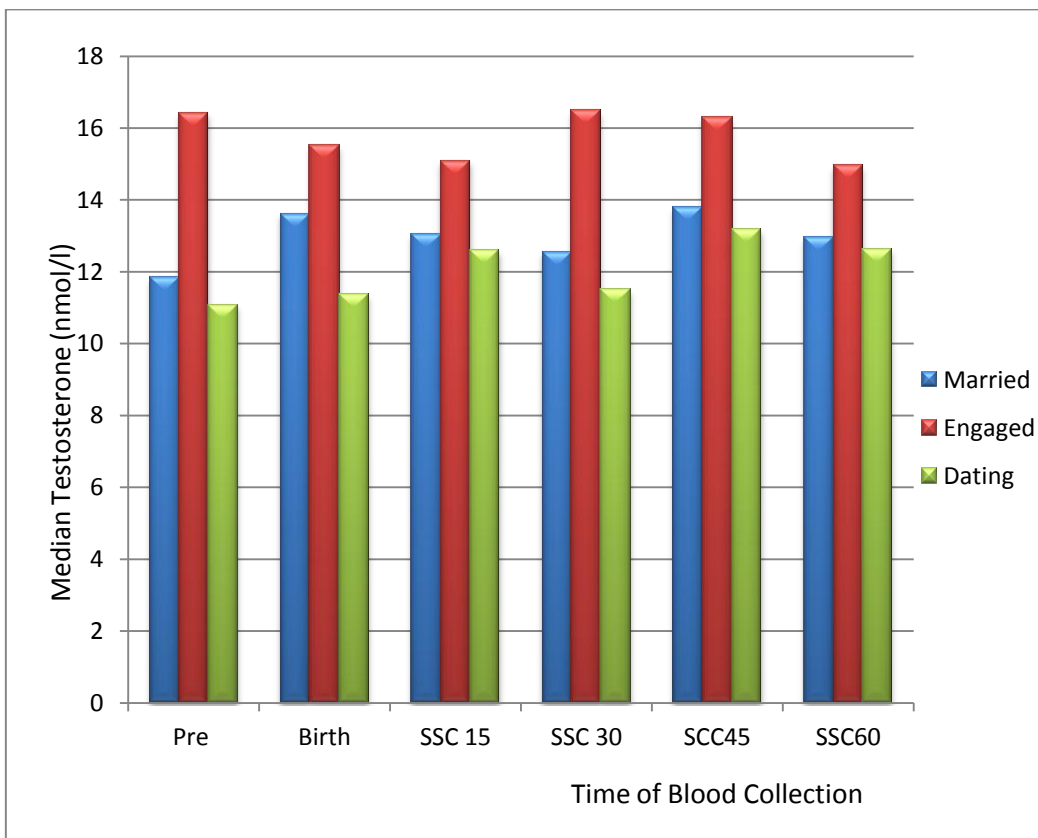


Figure 15: Testosterone trend of fathers grouped by relationship status

### 4.5.3 TESTOSTERONE VARIATION AND AGE

Figure 16 demonstrates the changes in the value of median testosterone with SSC of fathers aged 28 years and younger and 29 years and older. There is a large variation in the values of testosterone during SSC in both groups of fathers. At the time of the birth of the infant the median testosterone level of fathers aged 28 years and younger decreased from the base value ( $P=0.7$ ), after which it increased continuously until 45 minutes of SSC ( $P=0.6$ ), then dropped at 60 minutes of SSC with a  $P$  value of 0.4. At the time of the birth of the infant the median testosterone values of fathers aged 29 years and older showed a significant increase from the base value ( $P=0.04$ ). It then decreased continuously until 30 minutes of SSC ( $P=0.5$ ), increased at 45 minutes of SSC and dropped at 60 minutes of SSC in this group of fathers ( $P=0.2$ ).

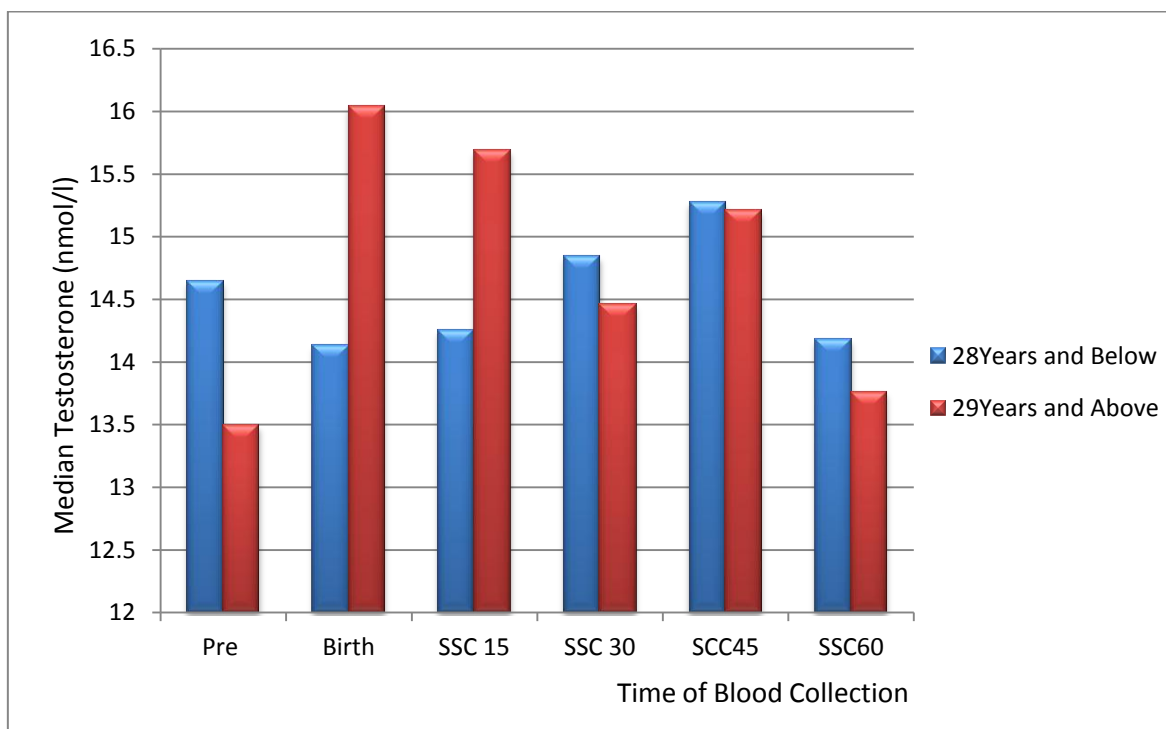


Figure 16: Testosterone trend of fathers grouped by age

## **4.6 SUMMARY**

This chapter presented the results obtained from analysis of the data, which included a non-parametric method of statistical analysis. Prolactin and testosterone levels were analysed separately, taking into consideration the effect of age, relationship status and type of C/S. The results show that in general prolactin levels decreased as SSC progressed, and that the levels of testosterone on the whole remained unchanged.

# CHAPTER 5: Discussion of results, implications and recommendations

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## 5 INTRODUCTION

This chapter discusses the results presented in chapter four. This study was designed to investigate changes in prolactin and testosterone levels during SSC between first-time fathers and their new born infants within six hours after birth. The hypothesis that prolactin levels would increase with SSC and that testosterone levels would decrease with SSC was tested.

### 5.1 CHANGES IN PROLACTIN LEVELS

At the time of the birth of the infant, the fathers' prolactin levels are seen to increase significantly ( $P=0.04$ ) (Figure 7). The increase at this point might have been due to the fathers hearing the sound of their infants' cry at birth. A study by Storey et al. (2000:184) showed that when first-time fathers were exposed to the cry of their infant immediately after birth, they experienced an increase in levels of prolactin. Fleming et al. (2002:406) also reported raised prolactin levels in fathers when they listened to the recorded sounds of infants crying, while non-fathers showed no change in prolactin levels. Comparing the values of prolactin from the time of first blood collection (point 1) until time at 60 minutes of SSC contact gives a P value of 0.2, showing that the decrease in prolactin level after 60 minutes of SSC is insignificant.

Generally prolactin levels are seen to decrease (Figure 7) from 15 minutes into SSC until 45 minutes of SSC ( $P= 0.004$ ); this may be due to the inhibitory control of dopamine, which is released from the hypothalamus due to the initial increase at the time of the birth of the infant (Grattan & Kokay, 2008:753). Brooks et al. (2005:362) and Almond et al. (2006:676) used a dopamine agonist such as bromocriptine and cabergoline to suppress prolactin in males, and observed that there was no change in paternal care before or after the suppression of prolactin. These results are not in keeping with those in other studies.

In a study by Roberts et al. (2001:217) prolactin levels were shown to be elevated after infant carrying by the paternally inexperienced common marmoset. Shradin et al. (2003:170) also demonstrated that prolactin levels were elevated one to three weeks after birth during infant carrying by paternally experienced *C. cupreus*, *C. jacchus* and *C. goeldii*. In these studies prolactin levels were not being tested immediately after birth but more than 24 hours thereafter. However, some studies report that prolactin levels decrease with increased contact time between father and child. Gettler et al. (2011:599) and Storey et al. (2011:356) have shown that fathers' prolactin levels drop from base line values after a period of 30 minutes play with their infants.

In this study prolactin appeared to increase between 45 minutes of SSC and 60 minutes of SSC; however, this increase was not significant as the Wilcoxon matched pairs test gives a P value of 0.5.

With respect to individual changes in prolactin values (Figure 6), at 15 minutes into SSC, nine fathers (P001, P003, P004, P005, P007, P008, P009, P010 and P011) experienced a decrease in the levels of prolactin, while two fathers (P002, P006) showed raised levels of prolactin. At 30 minutes of SSC, two fathers (P003 and P004) showed an increase in prolactin levels while prolactin levels dropped in fathers P002 and P006. The remaining fathers experienced a further decrease in prolactin levels. Fathers P007, P010 and P011 showed raised levels of prolactin at 45 minutes of SSC. At 60 minutes of SSC, seven fathers (P001, P003, P004, P005, P006, P007 and P010) showed an increase in prolactin levels, while four fathers showed a drop.

Prolactin levels fluctuated every 15 minutes in each individual father. Only one father (P009) demonstrated a progressive drop in the values of prolactin from the moment of first blood collection until 60 minutes of SSC. While it was observed that prolactin values fluctuated every 15 minutes at varying degrees in each of the fathers, the general trend seen is that prolactin levels decreased with SSC (Figure 7). The results of the study do not support the hypothesis that prolactin levels would increase with SSC between a first-time father and his child.

### **5. 1.1 EFFECT OF TYPE OF C/S ON PROLACTIN LEVELS**

The fathers were grouped into those whose infants were born by emergency C/S or elective C/S and were observed for changes in prolactin levels as SSC progressed (Figure 8). Fathers whose babies were born by emergency C/S had higher median prolactin levels throughout the process of SSC than those whose babies were born by elective C/S. The high prolactin values may be due to the fact that the father experienced an emergency situation which was unexpected and may have caused stress, which would trigger the release of high levels of prolactin (Euker, Meites & Riegler, 1975:185; Bodnar et al., 2004:208). Although prolactin levels are observed to increase at the time of birth and then decrease progressively, these changes are not significant. It is therefore concluded that the type of C/S performed has no significant effect on the changes in prolactin levels during SSC.

### **5.1.2 EFFECT OF RELATIONSHIP STATUS ON PROLACTIN LEVELS**

Fathers who were married to the mother of their child generally had higher levels of prolactin than those who were engaged or dating (Figure 9). Men who are married to the same partner for more than ten years have been shown to spend more time taking care of their children than divorced or single men (Ahmeduzzaman & Roopnarine, 1992:704; Amato & Sobolewski, 2001:900; Hofferth, 2006:53), and literature from non-human studies show that male animals who are in monogamous relationships show high levels of paternal care and have high prolactin levels (Ziegler & Snowdon, 2000:162).

### **5.1.3 EFFECT OF AGE ON PROLACTIN LEVELS**

Fathers were grouped according to their age and observed as to how age affected prolactin levels (Figure 10). Of the fathers who participated in the study, those who were older (29 years and above) were seen to have generally higher levels of prolactin than younger men (28 years and below). The higher levels of prolactin in older men may be due to the fact that prolactin increases in men with age (Vekemans & Robyn, 1975:738; Feldman et al., 2002:591). In both groups of fathers the median prolactin levels increased at the time of the infant's birth ( $P=0.2$  for fathers aged 28 and below and  $P=0.3$  for fathers aged 29 and above). It then decreased progressively up to 45 minutes

of SSC, after which it increased at 60 minutes of SSC, with P values of 0.6 for fathers aged 28 and below and 0.2 for fathers aged 29 and above. The P values for these changes are not significant. In this study age had no significant effect on changes in prolactin levels during the SSC process.

## **5.2 CHANGES IN TESTOSTERONE LEVELS**

The results showed that levels of testosterone in these fathers were not affected by the SSC process. Similar results were obtained in a non-human animal study by Hume and Wynne-Edwards (2005:306), where reduction in testosterone to levels below the lower limit did not have any effect on the retrieval and licking of pups by the dwarf hamster.

Some human studies have shown testosterone to decrease with paternal care. Gray et al. (2007:502) showed that testosterone levels in men dropped from baseline values after a 20 minute interaction between fathers and their toddlers. Gettler et al. (2011:16195) demonstrated that married men who were first-time fathers had a greater decline in morning and evening levels of testosterone compared to unmarried men with no children.

These results are also not in agreement with animal studies which suggest that testosterone promotes paternal care. Wang and De Vries (1993:156) showed that castration decreased paternal activity in Prairie voles which when treated with testosterone demonstrated paternal behaviour. Luis et al. (2009:437) study showed positive correlations between paternal care and levels of testosterone in the Volcano mouse.

With regards to individual changes in testosterone levels (Figure12) at the moment of birth, seven men (P003, P004, P005, P007, P008, P009 and P011) showed an increase in testosterone levels, while in four of the men (P001, P002, P006 and P010) testosterone levels decreased. At 15 minutes into SSC P007, P008 and P009 showed a continuous rise in testosterone, while P003, P004, P005 and P011 demonstrated a drop in testosterone. P001, P002 and P006 showed an increase in testosterone while P010 showed a continuous decrease.

At 30 minutes of SSC, six of the men (P002, P005, P007, P009, P010, and P011) demonstrated a decrease in testosterone levels, while the remaining five showed an increase in testosterone levels. Two of the men (P006 and P008) showed a progressive increase in testosterone levels throughout SSC.

Just as there was a great fluctuation in prolactin levels, testosterone values varied at each point of SSC in each individual; however, the general trend was that there was a minimal change in testosterone levels, suggesting that testosterone was not affected by SSC. The hypothesis that testosterone levels decreased with SSC was not supported.

### **5.2.1 EFFECT OF TYPE OF C/S ON TESTOSTERONE LEVELS**

Fathers whose infants were born by elective C/S were observed to have higher testosterone levels throughout the process of SSC than fathers whose infants were born by emergency C/S (Figure 14). The researcher could not find any explanation for this variation. It was also noted that the values of testosterone remained fairly constant throughout the SSC process in both groups of fathers. It was concluded that the type of C/S performed did not have a significant effect on the testosterone levels during SSC.

### **5.2.2 EFFECT OF RELATIONSHIP STATUS ON TESTOSTERONE LEVELS**

The study showed that men who were engaged to the mother of their children had the highest testosterone levels, followed by men who were married to the mother of their child (Figure 15). No explanation could be found for this variation. Also observed was that the values of testosterone remained fairly constant, with minimal changes throughout the SSC; hence relationship status has no significant effect on testosterone levels during the SSC process.

### **5.2.3 EFFECT OF AGE ON TESTOSTERONE LEVELS**

The study showed younger fathers to have higher levels of testosterone than older fathers at some points, while older men had higher testosterone values at other points (Figure 16). These changes are mostly insignificant, but a significant increase was observed in older fathers at the time of the

birth of their infant ( $P=0.04$ ). No explanation could be found for this increase, since testosterone levels are known to decrease with increasing age (Feldman et al., 2002:591).

### **5.3 IMPLICATIONS AND RECOMMENDATIONS**

This study showed that prolactin levels rose in fathers at the time when their infants were born and then drop during the process of SSC. The increase might have been due to the hearing of the sound of the infant crying. The infant's cry could act as an external stimulus which stimulates Prolactin-releasing factor from the neuroendocrine neurons which is released at the level of the median eminence. Prolactin-releasing factor is released into the long portal veins connected to the lactotrophs of the pituitary gland causing an increase in prolactin production and rise in blood levels (Reinhoffer et al., 2013:168). A rise in prolactin levels causes the activation of the neuroendocrine dopaminergic neurons (Grattan et al., 2008:498). Dopamine released from these neurons diffuses into the capillaries of the portal system and is transmitted to the anterior pituitary where it activates the D2 receptors on the lactotroph cells to inhibit prolactin secretions (Ben-Jonathan & Hnasko, 2001:741). Hence the drop in prolactin levels during SSC which may be due to the effect of the dopamine rise. This decrease of prolactin levels for the next one hour may be due to the breakdown of the released prolactin as its half life has been shown to be approximately  $40.8 \pm 13.8$  minutes (Yoshida et al, 1991:585).

It has been established that dopamine released in the hypothalamus due to high levels of prolactin causes a decrease in prolactin (Ben-Jonathan & Hnasko, 2001:724). Dopamine is known as the "reward circuit hormone", i.e. feelings of reward are associated with high levels of dopamine (Schultz, 2006:95), and it acts together with oxytocin to induce bonding (Liu & Wang, 2003:538; Aragona et al., 2003:3485; Swain, 2011:1249). The decrease in prolactin levels observed during SSC might be a result of the rising dopamine, which impacts on the bonding between the father and his infant, hence the father-child relationship could benefit from at least one hour of SSC soon after birth.

There were no significant changes in the levels of testosterone during SSC. The secretion of testosterone is regulated by feed-forward and feedback mechanism operating within the “hypothalamic-pituitary-gonadal axis” (Bhasin, 2008:646). GnRH released by the hypothalamus stimulates the release of LH and FSH which act on the Leydig cells to produce testosterone (Matsumo & Bremner, 1984:12). High levels of testosterone in the blood inhibit the release of LH by decreasing GnRH secretion and pituitary responsiveness to GnRH. According to Forester et al. (1997:3041) increase in LH concentrations in peripheral blood is preceded by a rise in the concentration of testosterone about 60 to 120 minutes later. A fall in LH would probably also cause a decrease in testosterone about 60 to 120 minutes later. It would probably therefore take some time before a decreased stimulation of testosterone levels would be reflected in plasma levels.

SSC not only encourages bonding between the infant and the parents, but is also beneficial for the health of the infant. The practice of SSC by fathers could therefore be encouraged in many communities, especially in low-income communities where there is a higher rate of infant morbidity and mortality. Education of fathers about this practice could be done by community health nurses during home visits to pregnant women as well as at the ante-natal clinics.

The intervention of SSC was conducted only for a period of one hour. Future studies could investigate SSC carried out for a period longer than just one hour and also observe prolactin and testosterone changes in the fathers for a period longer than this. One can postulate that there are changes which would be demonstrated in a study with a longer duration of SSC.

Paternal care has been shown to be important for the well-being of the child, and that early father-child bonding is an important factor for the future father-child relationship. It has been established that hormones have an impact on paternal care. The present study did not involve observing paternal behaviour and bonding during SSC. Future studies could observe paternal actions during SSC as well as have fathers completing a questionnaire on their feelings about the SSC experience. These behaviours could be compared with their hormone levels to better understand paternal care. Future studies could also involve follow-up visits at home to determine whether SSC immediately after birth has an effect on father-child interaction.

## 5.4 CONCLUSION

Parental care is important for child development and starts immediately after the child is born. Although some studies have shown that the influence a father has on his child is as important as that of the mother, it is not the same as that of the mother. It has been shown that hormones influence parental care, and that paternal care in humans has not been studied as much as maternal care.

This study was conducted to test the hypothesis that prolactin levels would increase during the process of SSC between first-time fathers and their newborns and that testosterone levels would decrease. However, the results of the study do not support these hypotheses. It was observed that prolactin levels decreased significantly during the process of SSC, while there was no significant change in the levels of testosterone during the SSC. Significant fluctuations in the hormone levels of each individual father were observed every 15 minutes.

An important finding in the present study is the significant increase in the levels of prolactin at the time of birth, which might be associated with fathers hearing the sound of their infants' first cry, and that the decrease in prolactin during SSC might be due to the rise of dopamine which reinforces the bond between father and infant.

In interpreting the results of this study the limitations of the study are taken into consideration. Time which is an extraneous variable could not be controlled given the fact that infants were born at different times of the day. This may have led to the variable hormonal levels in the fathers.

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## APPENDIX ONE

### ETHICS APPROVAL FORM



UNIVERSITY OF CAPE TOWN

Health Sciences Faculty  
Human Research Ethics Committee  
Room E52-24 Grootte Schuur Hospital Old Main Building  
Observatory 7925  
Telephone [021] 406 6626 • Facsimile [021] 406 6411  
e-mail: shuretta.thomas@uct.ac.za

21 September 2010

HREC REF: 429/2010

Ms NV Dzeayc  
c/o Ms H Barlow  
Division of Nursing and Midwifery

Dear Ms Dzeayc

**PROJECT TITLE: PROLACTIN AND TESTOSTERONE LEVELS IN FIRST TIME FATHERS WITH SKIN-TO-SKIN CONTACT WITH THEIR BABIES SOON AFTER BIRTH BY CAESAREAN SECTION.**

Thank you for submitting your study for review to the Faculty of Health Sciences Human Research Ethics Committee.

It is a pleasure to inform you that the Ethics Committee has **formally approved** the above-mentioned study.

**Approval is granted for one year till the 30<sup>th</sup> September 2011.**

Please submit an annual progress report if the research continues beyond the approval period. Please submit a brief summary of findings if you complete the study within the approval period so that we can close our file.

Please note that the ongoing ethical conduct of the study remains the responsibility of the principal investigator.

**Please quote the REC. REF in all your correspondence.**

Yours sincerely

**PROFESSOR M BLOCKMAN**  
**CHAIRPERSON, HSE HUMAN ETHICS**  
Federal Wide Assurance Number: FWA00001637.

S Thomas

## APPENDIX TWO

### INFORMATION SHEET

**Study Title: Prolactin and testosterone levels in first-time fathers during skin-to-skin contact with their infant soon after birth by caesarean section**

Dear father-to-be

I am a student at the University of Cape Town. I am doing a research on the effect that skin-to-skin contact with your new born infant has on the prolactin and testosterone hormones in you. The study is a requirement for my MSc degree in nursing

Hormones are your body's chemical messengers that travel through your blood stream to your heart, liver brain and other parts of your body. They affect processes such as growth, development, and sexual function. Prolactin is a hormone that is produced by the brain and its main function is that it stimulates milk production in mothers. Testosterone is a "male hormone" which is produced in the testes and its main function is that it encourages the development of male sex characteristics. Prolactin has been suggested to be a bonding hormone. There are other circumstances under which this hormone can be released and it lasts only for a short while in the blood stream. I want to find out if skin-to-skin contact with your infant will cause an increase in prolactin levels in your blood and decrease the level of testosterone.

Ethics approval has been obtained from the Faculty of Health Science Human Research Ethics Committee (REC REF: 429/2010). Permission to conduct the study has also been obtained from the medical superintendent of the department of Obstetrics and Gynaecology of the hospital.

## **Significance of the study**

The results of this study will increase our understanding of the associations of prolactin, testosterone and skin-to-skin contact in new fathers and may provide further evidence of the role of hormones in paternal care

The findings of the study would contribute to determine a scientific base for paternal care behaviours in human fathers as it would show that hormones are involved.

## **Procedure**

When you and your partner have agreed to take part in the study, you will both sign a consent form. I will meet you and your partner at the hospital on the day your partner is giving birth. The procedure will involve taking blood samples at a regular interval from a peripheral vein in your arm. An indwelling intravenous catheter will be inserted into your arm. It will be attached to a three-way stop-cock.

The first blood sample will be taken while your partner is being prepared for the operation. The second will be taken at the time when the infant is born. After the operation, your partner will be drowsy and may have some post-operative pain and she may not be able to take care of the infant. I would therefore like you to spend at least the first one-hour after delivery with your infant in skin-to-skin contact. This would be done after the paediatrician has examined the infant and confirmed that the infant is fit to be with its mother. A special shirt together with an inner cloth will be provided for you to wear over you and the infant that will keep the baby safe. You will have no shirt on and the infant will have only a nappy to prevent from wetting you.

The third blood sample will be collected after 15 minutes of SSC with the infant. The fourth, fifth and sixth blood samples will be collected every 15 minutes thereafter until the one hour of SSC is over. This means blood will be collected 6 times from you. The amount of blood collected each time

is 5 ml, which is equal to a teaspoon of blood. The whole process of doing the SSC process and collecting blood will take about two hours of your time.

Should you accept to participate in the study, you will not receive any money or other form of compensation for participating in the study. Your partner will still be given the full form of treatment and care should you refuse to take part in the study.

### **Discomfort/Risk**

You may feel some discomfort from the intravenous catheter inserted in your arm for collecting blood but this is temporary and will stop once the catheter is removed. To avoid pricking you several times, a three-way stop-cock will be used to open and close each time blood is being collected. There is a possibility of getting an infection from the site of needle insertion but this is very minimal. Measures to prevent infection such as using sterile material during blood collection, cleaning the area with alcohol swabs before inserting the catheter, and doing a clean dry dressing on the area when blood collection is completed will be taken.

### **Benefits**

There is no direct benefit to you if you participate in the study. However taking part in the study gives you an opportunity to bond with your new-born infant. The skin-to-skin process also helps to calm your infant, improve their breathing and stabilise its temperature. The results of the study may help increase our knowledge on the role of hormones in paternal care.

### **Ethical considerations**

Your acceptance to participate in the study is voluntary and you are free to withdraw at any moment before and during the study. Your partner must also be in agreement with this; as such her

consent is necessary for the study. All information obtained will be confidential and will not be shared with any other person not involved in the study without your permission. The results of the study will remain confidential and will be revealed as a number in the statistical analysis. Your privacy will be respected throughout the study. You and your partner will be put in a separate room during the procedure of skin-to-skin and blood collection and only I will have access to the room. You are free to ask any questions about the study at any time during the study.

If you would like to know about your results, you are free to contact me on the number 0766264118.

Researcher: Miss Ngah Veranyuy Dzeaye

Supervisors:

Mrs Hilary Barlow, Division of Child and Adolescent Health, UCT, Tell: 0216585723

Associate Professor Pat Mayers, Division of Nursing and Midwifery, UCT, Tell: 0214066464

Dr Nils Bergman, Department of Human Biology, UCT, Cell: 0824421079

UCT Health Sciences Faculty Human Research Ethics Committee

Room E52-24 Groote Schuur Hospital Old Main Building

Observatory

Tell. 0214066338

## APPENDIX THREE

### RESEARCH CONSENT FORM

**STUDY TITLE: Prolactin and testosterone levels in first-time fathers during skin-to-skin contact with their infant soon after birth by caesarean section**

The researcher has explained in detail to me about the study including the risks and benefits of the study. I have understood what the study is about. The researcher has made me understand that the intravenous catheter inserted may be uncomfortable but this is temporary and discomfort will stop when it is removed.

I agree to take part in the study. I have not been forced to participate. I do understand that if I choose to withdraw from the study at any time this will not have any effect on my family or me and my decision will not interfere in the care and support of my partner, my infant, or myself. I will not be compensated for taking part in the study. I have been given the opportunity to ask questions and all my questions have been answered.

The study will give me an opportunity to bond with my new-born infant. My partner/mother of the unborn infant has also given consent for me to participate in the study. I have been informed that ethics approval to conduct the study has been obtained from the Faculty of Health Science Human Research Ethics Committee (REC REF: 429/2010).

Father..... Sign..... Date.....

Mother..... Sign..... Date.....

Witnesses

Name..... Sign..... Date.....

Name..... Sign..... Date.....

## APPENDIX FOUR

### PERMISSION LETTER

Department of Health and Rehabilitation Sciences  
Faculty of Health Science  
University of Cape Town  
Observatory 7925

Hospital Medical Superintendent  
Hospital  
Observatory  
Cape Town

Dear Sir,

#### PERMISSION TO CONDUCT STUDY AT MATERNITY CENTRE

I am a postgraduate student at the University of Cape Town. I am doing a study on the effect of skin-to-skin contact between first-time fathers and their new-born infant within the first six hours after birth by caesarean section on the levels of serum prolactin and testosterone in the father. The study is a requirement for my MSc in Nursing.

The study involves obtaining peripheral blood samples of 5mls each six times from the research participants. Two will be taken before skin-to-skin and four during the process of skin-to skin every 15 minutes for the period of 1hour. Serum from the blood samples will then be analysed for the levels of prolactin and testosterone to determine if skin-to-skin has any effect on these hormones

I would be grateful if you permit me to do my study at the maternity unit of your Hospital. Ethics approval has been obtained from the Faculty of Health Sciences Research Ethics Committee, (REC REF: 429/2010)

Yours faithfully

Ngah Veranyuy Dzeaye

## APPENDIX FIVE

### DATA COLLECTION SHEET

**STUDY TITLE: Prolactin and testosterone levels in first-time fathers during skin-to-skin contact with their infant soon after birth by caesarean section**

Date of blood collection

Participant number.....

Demographic data

Age.....

Relationship status: Married..... Engaged..... Dating.....

Time of birth of infant.....

BLOOD SAMPLE	SAMPLE #	TIME
Taken before infant is born at time of preparation for C/S		
Taken at time of birth of infant		
Taken 15minutes after start of SSC		
Taken after 30 minutes of SSC		
Taken after 45 minutes of SSC		
Taken after 60 minutes of SSC		

