TRILOSTANE AS ANTIPROGESTIN THERAPY IN PREGNANCY TERMINATION

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DECLARATION

I, Philip M Zinn, hereby declare that the work on which this dissertation is based is my original work (except where the acknowledgements indicate otherwise) and that neither the whole work or any part of it has been, is being, or is to be submitted for any other degree in this or any other university.

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Abstract

Progesterone is central to the maintenance of pregnancy. Trilostane, a 3-beta-hydroxysteroid dehydrogenase inhibitor, was used to lower progesterone concentrations prior to prostaglandin administration in three clinical studies. 1) A comparative study of two trilostane dose regimens in the induction of midtrimester termination of pregnancy. 2) A placebo-controlled study of termination of pregnancy for fetal anomaly. 3) A placebo-controlled study of labour induction for intrauterine fetal death. End-points included the induction-to-delivery interval, alterations in placental and adrenal steroidogenesis and the acceptability of the treatment.

28 women requiring termination of pregnancy between 13 and 20 weeks gestation were recruited to the first study. A total dose of 720 mg of trilostane pre-treatment was as effective as 1080 mg of trilostane in reducing the interval to abortion after misoprostol. In the second study, 11 women requiring termination of pregnancy beyond 18 weeks gestation for serious fetal anomalies received either trilostane or placebo prior to misoprostol induction of labour. Although progesterone levels fell significantly with trilostane treatment, no reduction in the induction-to-delivery interval could be demonstrated. In the third study, 20 women presenting with intrauterine death were treated with either trilostane or placebo prior to induction of labour with prostaglandin E_2 and oxytocin. Six women delivered prior to the administration of prostaglandin. No benefit could be demonstrated for the use of trilostane in this clinical situation. In all three studies, adrenal function remained undisturbed during trilostane treatment and women found the therapy acceptable.

Trilostane is effective at low doses in the management of midtrimester TOP. The negative findings in the induction of labour studies may be influenced by small trial numbers, heterogeneity and inadequate medication dosages. Further studies and elucidation of the underlying physiology of labour in these different clinical situations is required.

INTRODUCTION

Termination of pregnancy

It may be required to end a pregnancy before the natural onset of spontaneous labour. Although a distinction is usually drawn between the induced abortion of a pre-viable fetus and induction of labour in the third trimester, methods that promote the various conditions favouring expulsion of the pregnancy and also the stimulation of uterine activity may be applicable to both situations.

The availability of family planning in a community depends on the interaction of geographic, economic, political, religious and social factors but, even when accessible, all currently available methods of contraception have an inherent failure rate. The demand for termination of pregnancy continues regardless of availability or legality and the morbidity and mortality of unsafe methods are well documented. It has been estimated that up to 53 million abortions are performed world-wide each year and approximately one-third are performed under unsafe conditions. The mortality due to unsafe abortion is estimated at 70 000 or 13% of the 500 000 maternal deaths each year.[1]

Vacuum aspiration is the most prevalent surgical method of pregnancy termination in the first trimester.[2] The procedure is safe when performed by experienced personnel but complications do occur. Serious morbidity may complicate up to 1% of cases and maternal deaths have been reported at rates of 2 women per 100 000. Minor morbidity, which includes psychological disturbances (serious psychological sequelae are rare), may be recorded in approximately 10% of women. Severe haemorrhage, infection, and genital tract trauma account for 85% of the 'major' complications and

prolonged uterine bleeding, retained products of conception or endometritis for most of the 'minor' morbidity.[2, 3, 4]

Second trimester termination of pregnancy is associated with a sharp and progressive rise in the complication rate and also of related costs. While comprising about 10-20% of all induced abortions, they are estimated to cause two thirds of major complications and half of the related maternal mortality.[5] The surgical technique of dilatation and evacuation has been shown to be both safe as a procedure in skilled hands and safer than induction of labour in early second trimester pregnancies.[6] In the recent past, induction of labour in the second trimester was usually achieved with prostaglandins alone – parenteral, extra- or intra-amniotic – and comparisons of the safety of surgical techniques with methods using a combination of antiprogestin and prostaglandin have yet to be made.

The gestational age at which women present for abortion is influenced by the availability and accessibility of services and by the provisions of the local abortion laws. Experience in European countries where 'liberalisation' of the law was accompanied by easy access to efficient services was a shift towards earlier abortion. In England and Wales, the proportion of abortions performed after the first trimester declined from 34% in 1969 to 14% in 1987.[7] The current experience at Groote Schuur Hospital, Cape Town, is that approximately 50% of women present for termination of pregnancy in the second trimester. Although 'liberalisation' of the law regarding termination of pregnancy in South Africa (1996) has removed barriers to safe abortion methods, late presentation may be a consequence of the inaccessibility or lack of services, poor referral systems, inadequate education and support structures or the problems of pregnancy denial and concealment.

Progesterone and uterine contractility

In a series of elegant experiments, Csapo originally described the suppressive effect of progesterone on uterine contractility.[8] Human studies subsequently confirmed that progesterone withdrawal in early pregnancy causes increased uterine contractility and leads to abortion.[9, 10] Progesterone appears to suppress both smooth muscle excitation and propagation. One mechanism may be the influence of progesterone on calcium binding to the cellular membrane. In addition, progesterone inhibits the formation of estradiol receptors and also of gap junctions, important for co-ordinated uterine activity.[11] Progesterone may also enhance the stability of lysosomal membranes. A reduction in local concentrations of progesterone may therefore facilitate the release of lysosomal phospholipase A_2, initiating increased prostaglandin production.[12]

Prostaglandins are potent uterotonic agents. In the normal menstrual cycle, under the influence of estradiol and progesterone, the endometrium acquires an increased capacity for prostaglandin synthesis. A high level of progesterone suppresses activation of these biosynthetic pathways in the luteal phase. Progesterone withdrawal due to luteolysis is followed by a release of prostaglandins from stromal and epithelial cells in the endometrium. In addition, the inhibition of prostaglandin dehydrogenase that is associated with a fall in progesterone concentrations further increases the effective local concentration of prostaglandins. Persistence of the corpus luteum in early pregnancy maintains progesterone levels preventing the release and suppressing the synthesis of prostaglandins by the decidua. The luteo-placental shift of progesterone production ensures continued high levels of circulating progesterone, maintaining a state of relative uterine quiescence until parturition.[13]

The importance of progesterone in the regulation of uterine contractility is supported by studies examining the effect of the progesterone receptor antagonist,

mifepristone,[14] and of progesterone synthesis inhibition with epostane[17] and trilostane[15] in early pregnancy.

Medical methods of pregnancy termination

Medical methods of pregnancy termination have advanced significantly over the past two decades. The combination of antiprogestins and prostaglandins has demonstrated greater efficacy than either compound alone in both the first and second trimester. The progesterone-receptor antagonist, mifepristone, and the 3β hydroxysteroid dehydrogenase inhibitors, epostane and trilostane, which decrease progesterone synthesis have been shown to successfully induce abortion in pregnancies up to 20 weeks gestation.[16, 17, 18, 19, 20] The incidence of complete abortion varies, however, from 85% at 6 weeks of amenorrhoea to 50% in gestations over 9 weeks. Prostaglandins, alone, are effective at inducing abortion but the high dose required is frequently associated with gastrointestinal side effects.

Misoprostol, a prostaglandin E_1 analogue, is currently the favoured compound. While having comparable efficacy to other prostaglandins in clinical use, it has the advantage of being relatively inexpensive and allows for easy storage and administration. Such properties offer a valuable alternative to other less stable and more expensive prostaglandin preparations, particularly in the context of developing countries. Used alone, over 50% of women in the first trimester will require more than one dose. When administered 36-48 hours after mifepristone, 95% of pregnancies up to 9 weeks gestation will abort completely within 8 hours following a single dose of misoprostol (800 μg misoprostol vaginally).[21] In most women, abortion occurs within 4 hours.

In studies of second trimester termination of pregnancy with misoprostol alone, success rates vary from 73 – 92 %. These follow single doses of misoprostol, 100 to 800 μg vaginally, and report an average induction-to-abortion interval varying from 11 to 28 hours.[22] Bugalho *et al*[23] reported an 80% success rate, within 18 hours, following a single dose of misoprostol 800 μg vaginally. Two further doses of 400 μg, at 18 and 24 hours respectively, increased the success rate to 89%.

Although developed for oral ingestion, vaginal administration of misoprostol appears to be superior to the oral route in both first and second trimester studies.[21, 24] This would seem to be due to differences in pharmacokinetics: oral administration produces a high peak and steep fall over 120 minutes whereas vaginal administration demonstrates a more gradual rise and fall, hence a longer duration of action.[25]

Studies by El Rafaey and colleagues, combining misoprostol and mifepristone, reported success rates of 90%[26] (oral misoprostol 400 μg/3h, maximum 1200 μg) and 97%[27] (misoprostol 600 μg vaginally + 400 μg orally or vaginally 3h later) with initiation of prostaglandin treatment to abortion intervals of 8h and 6.4h respectively.

A placebo-controlled study investigating the use of trilostane in second trimester termination was conducted previously at Groote Schuur Hospital, University of Cape Town.[28] Following 48-72h of oral trilostane, misoprostol (200 μg/4h) was administered vaginally. A significant reduction of 57% in mean prostaglandin to abortion interval was observed in the trilostane group (9.2h) compared to placebo (21.6h). In addition, a reduction in pain, analgesia and total misoprostol doses was demonstrated in the treatment group. Importantly, there was no adverse effect on adrenal steroidogenesis.

The benefits of progesterone inhibition to facilitate medical termination of pregnancy are well established. Due to licensing regulations and cost, however, the availability

of mifepristone is naturally limited, particularly in the developing world. Although soon to be licensed in South Africa, the cost of mifepristone is likely to remain an important factor determining its role in general practice. For many other countries, pharmaco-political and economic considerations suggest that it may not become readily available in the near future.

As mentioned above, clinical studies in human pregnancy using the 3-β-hydroxysteroid dehydrogenase inhibitors, epostane and trilostane, received initial attention in the 1980s. A decision by the pharmaceutical company not to pursue this field of investigation at the time, however, led to withdrawal of these compounds from clinical interceptive research. Recent acquisition of the development rights to trilostane by Stegram Pharmaceuticals Ltd (Billingshurst, Sussex, UK) has again made this drug available.

Termination of pregnancy for fetal anomaly

With the development of ultrasound technology, screening techniques using maternal serum markers for fetal anomaly and methods of prenatal diagnosis in fetal assessment, the need to terminate a pregnancy because of a serious fetal anomaly has become more frequent. Current imaging and diagnostic methods to detect fetal anomaly are typically undertaken in the second trimester and the decision to terminate an affected pregnancy usually follows some weeks of review, frequently beyond 20 weeks gestation. Serious fetal anomalies are also diagnosed later in pregnancy during the investigation of abnormal clinical findings.

The methods of prostaglandin termination of pregnancy described above are usually applied to these circumstances. The safety of these methods, however, in later gestation when uterine sensitivity to oxytocic agents is naturally increasing, is not

clear and modification of drug regimens for different gestations has been largely empirical. Furthermore, the practice of fetocide prior to termination of pregnancy beyond 20 weeks gestation is not universal and the response to methods of termination may differ in the presence or absence of an intact fetoplacental unit.

Fetal anomalies account for the majority of pregnancy terminations after 18 weeks while termination of pregnancy for reasons of serious maternal risk contribute most of the balance. Termination of pregnancy for serious fetal anomalies after 18 weeks gestation therefore provides an opportunity for the development of gestation-specific dosage regimens and may also offer insights that enable improvement of induction of labour in the third trimester.

Management of intrauterine fetal demise

Historically, fetal demise was managed by watchful waiting in the knowledge that spontaneous labour would usually ensue within a few weeks. Frequently, due to a lack of reliable methods for the diagnosis of fetal death, confirmation of this was only possible at the time of delivery. Dippel, in a review of 306 intrauterine deaths beyond 28 weeks gestation, reported that 75% laboured spontaneously within 14 days and 89% within 21 days.[29] In contrast, Tricomi, reviewing 165 cases of intrauterine death in a cohort of infants weighing over 1000g, reported a 90% and 93% delivery rate within 14 and 21 days respectively.[30] Differences between the groups may be explained by the high proportion of women with placental abruption (32%) and patients managed by caesarean section (12%) or induction of labour (11%) in Tricomi's group. The intrauterine deaths in Dippel's group were more frequently associated with syphilis.[31]

Some women do not labour spontaneously for many weeks after fetal demise. With the advent of more rapid and reliable means of diagnosis, however, the need to procure delivery soon is often motivated by the degree of emotional distress. The risks of intrauterine sepsis and consumptive coagulopathy are also cited as indications for induction[32] although, in the absence of massive placental abruption, maternal hypofibrinogenaemia is uncommon in the first month after fetal demise as is infection in the presence of intact membranes. Observations by Prichard[33] suggested that plasma fibrinogen levels decline after 4 weeks, at a steady rate of 0.2-0.85 g/l per week. In his series of over 100 women, a level below 1.5 g/l was not observed less than 5 weeks after the presumed date of fetal demise. A fibrinogen level above 1 g/l is required for blood coagulation. The use of heparin to inhibit the intravascular coagulation process that causes the hypofibrinogenaemia allows spontaneous correction of plasma fibrinogen levels but the availability of fresh frozen plasma has generally superseded this approach in modern obstetrics.

Prostaglandins were first used for the management of patients with intrauterine fetal death in 1970.[34] Since then, many authors have reported on the use of various prostaglandin preparations by different routes and regimens. All studies were uncontrolled and protocols frequently included the use of oxytocin infusions. Initially prostaglandin E_2 (PGE_2) was used intravenously (up to 5 μg/min) and then vaginally with the manufacture of 20 mg pessaries. Prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$), intramuscular and intra-amniotic, produced similar results to PGE_2. Further manufacturing developments produced PGE_2 gel, used as an extra-amniotic and intracervical preparation, and misoprostol (a PGE_1 analogue). Where third trimester subgroups could be distinguished in these studies, mean induction-to-delivery intervals ranged between 7 and 18 hours.

Gordon and Pipe[35] made a comparison of prostaglandin and oxytocin for the management of intrauterine death. They obtained a mean induction-to-delivery interval of 11.08h with PGE_2 intravenously and 22.42h using oxytocin incrementally up to 2400 mu/min. In both arms of the study the failure rate was 20% (undelivered within 24 hours, n=15 in each group) however those in the prostaglandin group delivered soon after recommencing the infusion while the oxytocin-treated patients required a much longer duration of therapy. In general, most studies report a failure of treatment, usually defined as failure to deliver within 24-36 hours, of between 3% and 7%.

In studies where PGE_2 has been administered vaginally, total doses have frequently been in excess of 40mg.[36, 37, 38] Although rare, serious complications have been reported. Uterine rupture has been documented,[39] particularly in association with a scarred uterus and when combining prostaglandin and oxytocin therapy.[40] Tonic uterine contractions may occur in late third trimester prostaglandin induction and cervical tearing, as a consequence of precipitate labour, can necessitate hysterectomy.[36, 41] Large doses of vaginal prostaglandin E_2 have also been associated with myocardial infarction.[42]

All studies report a high incidence of side effects, particularly gastrointestinal and thermoregulatory. These are well-recognised problems of prostaglandin use, the severity of which seems dose-related, and includes nausea and vomiting (20-81%), diarrhoea (7-77%), pyrexia (5-44%) and rigors (up to 12%). A summary of the studies utilising prostaglandin regimens in the management of intrauterine death is presented in Table 1.

Investigation of the role of antiprogestins in the management of intrauterine fetal death (IUD) has a number of potential benefits. An improvement in the efficiency of

pregnancy termination in the presence of fetal demise is required. The wide range of induction-to-delivery intervals, the failure rates and complications of over-stimulation described above illustrate the unpredictability of the uterine response to prostaglandins. The persistence of placental activity following non-placental causes of fetal death is appreciated albeit poorly understood (and there is a lack of longitudinal data in this field).[51] With continuing progesterone production by the placenta, it could be hypothesised that progesterone inhibition may assist in pregnancy termination after fetal demise in a similar fashion to that observed with earlier viable pregnancies. If so, the benefits may include a greater predictability and a reduction of the induction-to-delivery interval, reduced prostaglandin and analgesia requirements, less side effects (lower prostaglandin doses) and a lower complication and failure rate of prostaglandin labour induction. These benefits could apply, to some extent, to induction of labour with a viable fetus however, due to the unknown fetal effects of these medications, an assessment of antiprogestins in the third trimester without fetal risk is important. In addition, our understanding of mechanisms of cervical ripening and of the onset of labour could benefit from such research.

Selinger[43] used epostane in 7 women with intrauterine fetal death between 18 and 41 weeks gestation (mean gestational age of 24 weeks). After 24 hours of treatment with epostane, prostaglandin E2 gel was administered vaginally as a single 25mg dose. Oxytocin (100 mu/min) was commenced 15-20 hours later if required. Using historical controls (n=4), he could not demonstrate a significant difference in induction-to-delivery interval between the two groups (epostane – 6.9h, SD 2.4h; control – 12.3h, SD 8.6h). As the sample size is small,[44] a conclusion cannot be drawn in this respect. A significant fall in progesterone levels in the treatment group was demonstrated.

Cabrol *et al,*[45] in an uncontrolled study, used mifepristone in two dose regimens for two or three days in 18 women with intrauterine death. The mean gestational age was 24 weeks (18-38). Prostaglandin or oxytocin was commenced after 72 hours if delivery had not yet occurred. At 200 mg twice daily, 9 of 11 women delivered within a mean time of 39 hours (range 20-58 h). In a lower dose regimen, 100 mg daily, only 2 of 7 women delivered within 72 hours. They concluded that a dose-related labour-inducing effect of mifepristone may be present. Padayachee *et al,*[46] in a randomised controlled trial, used mifepristone (200 mg twice daily) or placebo for a maximum of 3 days in 24 women with intrauterine fetal death. All were beyond 20 weeks gestation with a mean gestational age of 32 (SD 1) weeks. No additional therapy was given in this 72 hour period. After 72 hours, significantly more women had delivered in the mifepristone group (n=8) compared with placebo (n=2), P <0.02, although intervals to delivery and subsequent response to oxytocics were not reported.

Table 1. Studies of induction of labour with prostaglandin in the presence of fetal demise.

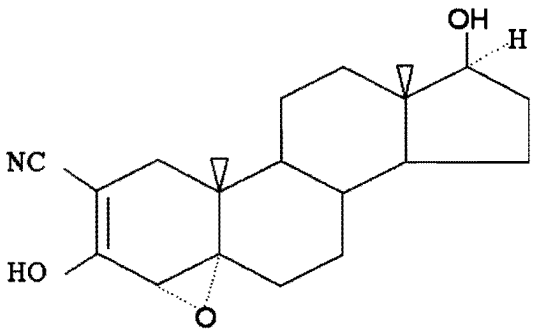
Investigator (first author only)	Number (n) (3rd trimester only)*	Gestational age (weeks)§	Protocol	ID Interval (hours)§
Karim[34] 1970	15	34.7	PGE_2 0.5-2.0 µg/min IV	12.17
Filshie[47] 1971	10	31.7	PGE_2 0.5-2.5 µg/min IV	9.82 (7.2-12.5)
Naismith[48] 1974	17	33	PGE_2 0.5-1.0 µg/min IV + oxytocin 2–1024 mu/min	7.42 (2.3-21)
Gordon[35] 1975	15	29	PGE_2 0.625-5 µg/min IV	11.08
Bailey[36] 1975	13	33.8	PGE_2 10-20mg PV q3-8h	6.9
Kent[37] 1976	19	27.5	PGE_2 20mg PVq3-4h + oxytocin 10-20 u/l if SRM	7.9 (2-17)
Ylikorkala[49] 1976	6	30.7	$PGF_{2\alpha}$ 100-250 µg IM q2-4h	8.57 (4-20)
Karim[50] 1976	26	34.7	$PGF_{2\alpha}$ 0.5 mg IM q8h	11.07 (2.8-32)
Southern[42] 1978	709	26.8	PGE_2 20mg PV q3-5h	10.7
Sher[51] 1979	20	32	$PGF_2\alpha$ 30mg + urea 60g IA	12.4 (2-24)
MacKenzie[32] 1979	23	36.2	PGE_2 5mg PV x 1 + oxytocin 4 -32 mu/min	14.7 (4 0 – 33.5)
Lauersen[52] 1980	26	31.6	PgE_2 20mg PV q2-3h ± oxytocin up to 16mu/min	9.56
Scher[38] 1980	26 23	31 26	PGE_2 375 µg EA q2h PGE_2 20 mg PV q3-6h	8.6 9.2
Karim[53] 1982	282	>24	$PGF_{2\alpha}$ 0.5mg IM q8h	10.5
Rath[54] 1985	22 20	33.7 33.1	$PGF_{2\alpha}$ 5mg IC then EA PGE_2 0.5-1mg IC then EA	18.1 (4.8-36) 13.7 (3-26)
Kent[41] 1986	46	28-45	PGE_2 10mg PV q2-4h	8.7 (2-19)

*Where a subgroup could be defined, only third trimester figures are reported.

§ Reported as means.

IA Interval-induction-to-delivery interval, IV-intravenous, IM-intramuscular, IC-intracervical, IA-intra-amniotic, EA-extra-amniotic, PV-vaginal, SRM-spontaneous rupture of membranes.

Trilostane



Trilostane (4α 5 epoxy – 17β hydroxy – 3 oxo – 5α androstane – 2α carbonitrile) selectively and reversibly inhibits the enzyme system - 3β hydroxysteroid dehydrogenase Δ5 oxosteroid isomerase (3βHSD) – essential for the synthesis of all steroids in the adrenal glands, ovaries, testes, and placenta. It was developed in the 1970s by Sterling-Winthrop Research and Development, Guildford, Surrey, UK as WIN 24 540, along with two similar compounds, epostane (WIN 32 729) and azastene (WIN 17 625).

Trilostane is currently licensed and marketed in the United Kingdom by Sanofi-Winthrop for the management of adrenal cortical hyperfunction and post-menopausal breast cancer. In the management of cortisol or aldosterone excess, it lowers both glucocorticoid and mineralocorticoid steroids, however, normal feedback mechanisms in the hypothalamo-pituitary-adrenal (HPA) axis and the renin-angiotensin-aldosterone (RAA) system prevent excessive lowering of cortisol and aldosterone respectively. Trilostane reduces progesterone levels by inhibiting the essential conversion of pregnenolone to progesterone by the 3βHSD enzyme complex (see diagram, page 33). Pregnenolone, which increases dramatically in concentration as a consequence of 3βHSD inhibition, is biologically inert. Trilostane has no agonist or antagonist activity to estrogen, progesterone or androgen receptors and does not

inhibit aromatase activity. Its effect on sex steroid activity is thus solely due to an inhibition of steroid biosynthesis.

In the treatment of adrenal cortical hyperfunction, the dosage ranges from 120-480 mg/day but may be increased to 960 mg/day. For postmenopausal breast cancer, the maintenance dose is 960 mg/day (or 720 mg/day if the higher dose is not tolerated) combined with a physiological replacement dose of hydrocortisone (e.g. 30 mg /day in divided doses).

Common side effects - with reported frequencies in some studies[28, 55, 56] - include nausea and vomiting (30-37%), flushing, tingling and swelling of the mouth (10-13%) and mild diarrhoea (17-50%). The facial symptoms are not an allergic response and do not necessitate cessation of therapy. Rhinorrhoea and rashes are less common. Granulocytopenia has been reported but is rare (and reversible), typically associated with bone marrow compromise due to disease or chemotherapy.

Epostane acts similarly to trilostane and was originally thought to be more potent as an interceptive agent than as an inhibitor of adrenal steroidogenesis. While trilostane is derived from testosterone, epostane is a 17α-methyl-testosterone derivative. Epostane has not been developed for clinical use and, together with trilostane, was withdrawn from clinical investigation into its interceptive properties in the 1980s without these having been fully explored. The licence for the further development of trilostane is now held by Stegram Pharmaceuticals Limited (Billingshurst, Sussex, UK) where an interest in its development for gynaecological indications have made it available once again.

The potential of trilostane as an interceptive agent hinges on its ability to suppress progesterone synthesis in the adrenal, ovary and placenta. This anti-progesterone quality may have clinical value for both fertility and pregnancy regulation. With

evidence for a critical role of progesterone in the process of ovulation,[57] inhibition of progesterone at this time may prove contraceptive. Similarly progesterone inhibition can disrupt the luteal phase, preventing implantation.[58, 59, 60] The use of antiprogestins in termination of pregnancy has already been mentioned (page 18) and a well-documented potential for cervical ripening has implications for cervical preparation prior to surgery or induction of labour.[61] Most research in this field has been conducted with mifepristone (RU486) and the potential for progesterone synthesis inhibitors to behave similarly has yet to be realised.

Studies of induction of abortion and labour using trilostane

Three studies, investigating the role of progesterone synthesis inhibition with trilostane in abortion and induction of labour, form the basis of this dissertation. Mifepristone is not currently available in South Africa or in other developing countries and, although it has proven efficacy in first and second trimester termination of pregnancy, its cost may well be a limitation if it were available. The efficacy of progesterone synthesis inhibition with trilostane in termination of pregnancy has already been demonstrated and may present an alternative strategy to progesterone receptor blockade.[20, 28]

The first study assesses the efficacy of lower doses of trilostane, in comparison to earlier work from this department, in midtrimester termination of pregnancy. Any reduction in dosage requirements could be beneficial in reducing the incidence of dose-related side effects and medication costs but should not compromise efficacy. Two other studies investigate the potential of trilostane to reduce the induction-to-delivery interval in pregnancies beyond 18 weeks. Women requiring termination of

pregnancy for serious fetal anomalies and women presenting with intrauterine fetal death beyond 26 weeks gestation were recruited for these studies respectively.

Misoprostol

In 1973, G.D. Searle & Co. (Chicago) developed misoprostol, a prostaglandin E_1 analogue, for the treatment of NSAID-induced peptic ulcer. It's uterotonic and cervical ripening qualities had been appreciated and were reported on by Rabe *et al*[62] in 1987 and Edwards *et al*[63] in 1994, respectively. Consequently, misoprostol has become widely used in termination of pregnancy (TOP) at all gestations although this remains an unlicensed application.

The hydroxypropyl methylcellulose base confers stability at room temperature for several years and it is relatively inexpensive. This makes it an attractive alternative to prostaglandin preparations such as dinoprostone and gemeprost (which are expensive and require refrigeration), particularly in developing countries.

Studies of misoprostol used as a single agent in early TOP have generally not produced promising results, mostly utilising 200 to 800 μg as a single vaginal dose. Success rates may improve with a multiple high dose regimen and in the second trimester. The success of misoprostol in medical TOP is significantly enhanced when combined with mifepristone or methotrexate.[64]

With respect to efficacy and side effects, vaginal administration seems preferable to the oral route where gastrointestinal symptoms (nausea, vomiting and diarrhoea) are more common. Oral administration results in a high peak followed by a steep fall of serum concentrations within 120 minutes. A more gradual rise and fall is seen with vaginal administration, hence a longer duration of action.[65] In Ho's study,[24] comparing vaginal and oral misoprostol (after mifepristone) in second trimester TOP, the greater

efficacy for the vaginal route was supported but the women themselves preferred the oral route.

Dinoprostone

Dinoprostone, a synthetic analogue of prostaglandin E_2, is commonly used for induction of labour. Its effect on smooth muscle results in the stimulation of uterine contractions as well as dose-related side effects including nausea, vomiting, abdominal cramping and diarrhoea. It acts as a vasodilator and may be associated with modest hypotension. The effect of cervical ripening (softening, effacement, dilatation) is incompletely understood but possible mechanisms include an increase in collagen breakdown or an alteration in collagen binding and tissue hydration by altering glycosaminoglycans (GAG) and proteoglycan composition.[66]

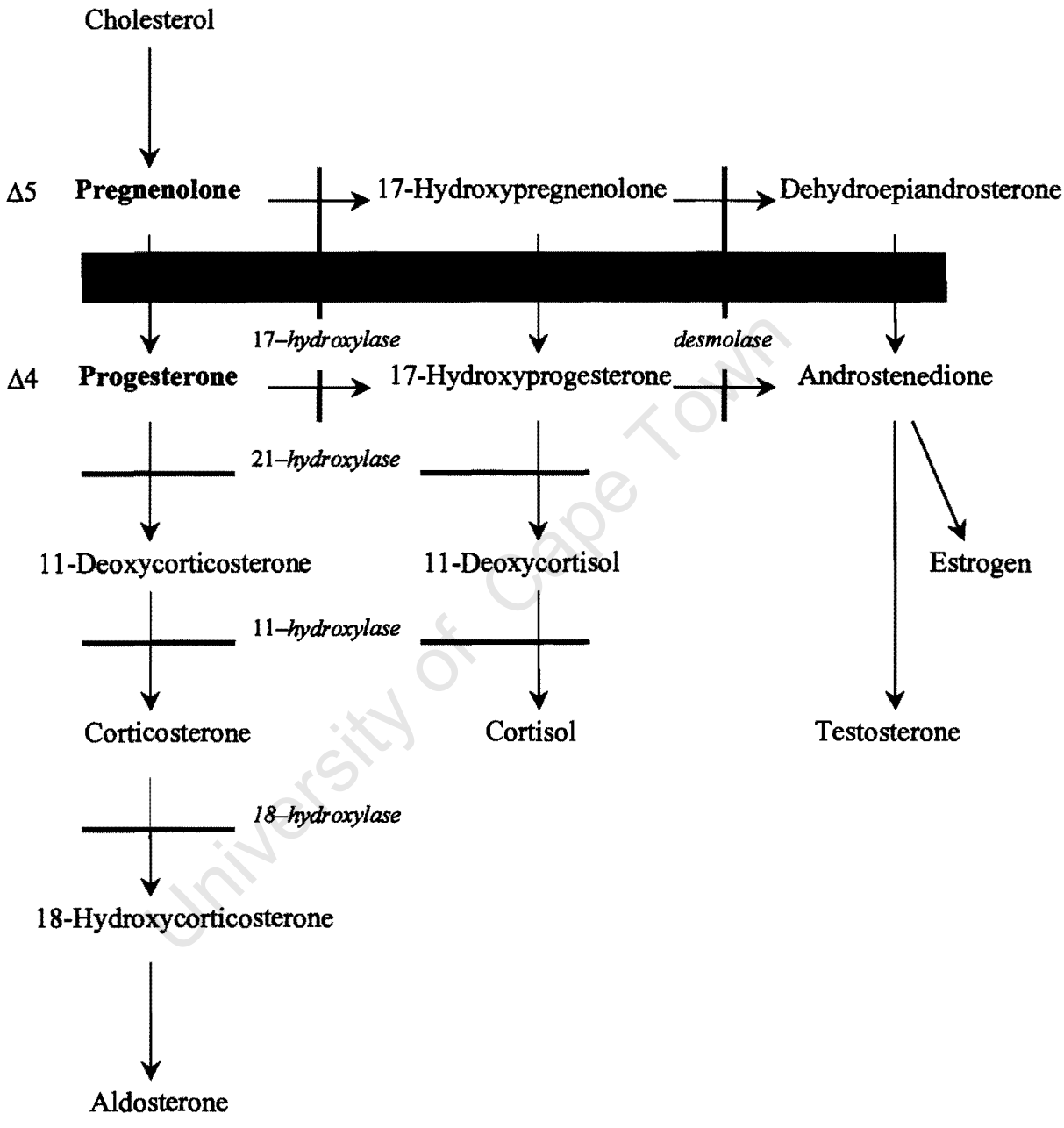
The vaginal gel preparation contains dinoprostone, 1 or 2 mg / 3 g (2.5 ml), in colloidal silicon dioxide and triacetin gel. It is packaged in a single-dose syringe system designed to deliver 1 or 2 mg of active ingredient into the posterior vaginal fornix. It requires refrigerated storage (2-8°C).

Oxytocin

In 1906 Sir Henry Dale demonstrated that intravenous injections of extracts of posterior pituitary gland caused uterine contractions in the gravid cat. Vincent du Vigneaud identified the hormones of the posterior pituitary and synthesised oxytocin in 1953, receiving the Nobel Prize for his work. Oxytocin is a neuropeptide with nine amino acid residues and a ring structure essential for its biological activity. Very similar to vasopressin (anti-diuretic hormone), it displays some vasoactive and anti-diuretic effects when used in large doses. Circulating oxytocinase, produced by the placenta, destroys the ring structure of oxytocin, eliminating its biological activity.

The plasma half-life of oxytocin is short, from 5 to 17 minutes.[67, 68] Apart from a direct uterotonic effect, oxytocin stimulates production and release of arachidonic acid and $PGF_{2\alpha}$ from the decidua potentiating uterine contractions. Myometrial responsiveness to oxytocin begins at about 20 weeks and increases gradually to 30 weeks and then markedly increases to a maximum in spontaneous labour at full term. This is reflected by increasing myometrial and decidual oxytocin receptors, reaching a maximum after the onset of labour (whether term or preterm). There is a fetal component to the circulating maternal oxytocin concentrations in labour. Concentrations consistent with that achieved naturally in the first stage of labour may be achieved with intravenous infusion rates of from 2 to 6 mu/min.[69] A higher infusion rate is generally required for induction of labour, compared to labour augmentation, with 30% of induced patients requiring more than 20 mu/min compared to 3% of augmented patients.[70]

STEROID BIOSYNTHETIC PATHWAYS



PATIENTS AND METHODS

Introduction

Trilostane was administered prior to termination of pregnancy with prostaglandin in three clinical trials.

Study 1 – Midtrimester termination of pregnancy

Study 2 – Termination of pregnancy for fetal anomaly

Study 3 – Induction of labour for fetal demise

The aim of these trials was to assess the efficacy of trilostane in reducing the interval to delivery after prostaglandin administration.

The midtrimester termination of pregnancy trial was based on, and was supplementary to, a placebo-controlled trial conducted previously by members of our research group.[28] The efficacy of lower total trilostane doses (relative to the placebo-controlled trial) was assessed.

The effect of trilostane in pregnancy termination for fetal anomaly and induction of labour for fetal demise was assessed by comparison with placebo.

In all of the studies, the primary outcome was the induction-to-delivery interval (induction = first administration of prostaglandin). Secondary outcomes included side effects, pain, patient satisfaction and acceptability of the procedures (global satisfaction) and effects on adrenal and placental steroidogenesis.

Study Design

The study groups were drawn from women attending the teaching hospitals of the Faculty of Health Sciences of the University of Cape Town during 1998 and 1999. Women were eligible for recruitment to the studies in the following circumstances.

Study 1. Women requiring termination of pregnancy between 13 and 20 weeks gestation.

Study 2. Women requiring termination of pregnancy after 18 weeks gestation for reasons of fetal anomaly.

Study 3. Patients requiring termination of pregnancy for intrauterine death after 26 weeks gestation.

The following exclusion criteria applied in all 3 studies:

1. Inability to consent due to age (under 18 years)
2. A lack of understanding of the study or communication difficulties
3. Presence of a multiple pregnancy or a scarred uterus
4. Existence of a medical or obstetric condition where delay in delivery would be unwise or for which the use of trilostane, prostaglandin or oxytocin might be contraindicated.

Study 1. Midtrimester termination of pregnancy

Patients were recruited to this study at the Groote Schuur Hospital, which is the tertiary teaching hospital for the University of Cape Town. Patients seeking termination of pregnancy (TOP) in this region are typically referred from a primary health care service to the hospital serving their catchment area. At Groote Schuur Hospital, in a dedicated clinic, assessment according to the Choice on Termination of

Pregnancy Act, No 92 of 1996, (see Appendix) and counselling precedes the decision to terminate the pregnancy. Second trimester (13-20 weeks gestation) pregnancy termination is conducted on an inpatient basis in the general gynaecology ward, usually within one week of the clinic assessment. The standard method of termination is medical, using misoprostol. This is administered as an initial dose of 400-600 μg vaginally followed by 200-400 μg doses orally or vaginally, 3-4 hourly as required.

Patients were recruited to the trial at the time of their admission to the gynaecology ward. A full explanation of the standard method of medical TOP and the purpose and method of the trial was given. Patients not wishing to participate in, or excluded from, the trial continued with TOP in the standard manner. Patients who gave consent (see *Consent*) commenced trial medication on the following day.

Patients were randomised (see *Randomisation*) to receive a total trilostane dose of either 720 mg (Group 1) or 1080 mg (Group 2) over 48 hours. Doses were divided into twice or three times daily administration depending on the regimen and were incremental (e.g. Day 1 – 120 mg bd, Day 2 – 240 mg bd). Treatment with prostaglandin E_1 (misoprostol) - was commenced on the day following completion of the trilostane course. A single misoprostol tablet (200 μg) was administered vaginally every four hours until abortion was effected, to a maximum of six doses. If unsuccessful after 24 hours the induction was considered a failure and further management was determined by the clinical team as appropriate to the individual. (See diagram, page 42.)

Study 2. Termination of pregnancy for fetal anomaly

Patients were recruited to this trial at Groote Schuur Hospital having attended a clinic of the Fetal Anomaly Group where a decision to terminate the pregnancy on the

grounds of a severe fetal anomaly had been made. Patients are referred from throughout the Western Cape Province for tertiary level ultrasound assessment, clinical genetic counselling and prenatal diagnosis. Where appropriate, patients are offered termination of pregnancy. Once a decision to proceed with TOP had been taken, patients were counselled about the methods of TOP and invited to take part in the clinical trial. The standard procedure for TOP, in these circumstances, would involve elective admission to an antenatal ward within one week of the decision. Prostaglandin induction of labour (misoprostol) would normally be commenced, alternative methods would be considered if unsuccessful or contra-indicated. Patients consenting to the trial followed the same procedure for admission and were then randomised to receive either trilostane or placebo for two days prior to commencement of misoprostol.

A total dose of 720 mg of trilostane was administered, 120 mg twice-daily on Day 1 followed by 240 mg twice-daily on Day 2. Misoprostol was administered vaginally every four hours, commencing on Day 3, until uterine evacuation was effected. A regimen of two 100 μg doses (one half of a cleaved 200 μg tablet) followed by two 200 μg doses was used. The trial protocol ceased at delivery or 24 hours after commencing induction with prostaglandin. If undelivered at this time, further management was determined on an individual basis by the clinical team. (See diagram, page 42.)

Study 3. Induction of labour for intrauterine fetal death

This trial was conducted at the Groote Schuur Hospital. Patients were recruited from the three teaching hospitals (including Groote Schuur Hospital) attached to the University of Cape Town. Once a diagnosis of intrauterine fetal demise (IUD) had

been made, the patient was counselled about further management. The standard procedure would involve induction of labour with prostaglandin and /or oxytocin as appropriate. This would take place electively (usually commencing within 24 hours) unless urgent delivery was indicated for maternal reasons or induction of labour was contra-indicated. Patients were counselled and invited to take part in the trial, usually commencing the following day, with transfer to Groote Schuur Hospital if required. Consent was obtained followed by randomisation to receive either trilostane or placebo for two days prior to the administration of dinoprostone gel.

The trilostane regimen was the same as has been described for Study 2. On Day 3, labour was induced with dinoprostone gel vaginally, repeated after four hours if required. An intravenous oxytocin infusion was commenced four hours after the second dose of gel if labour was not yet established. Oxytocin was commenced at 2.5 mu/min, increasing by 2.5 mu increments every 30 minutes to a maximum of 20 mu/min. The infusion was stopped after 14 hours if labour was not established (see diagram, page 42). The trial protocol ceased at delivery or 24 hours after commencing induction with prostaglandin. If undelivered at this time, further management was determined on an individual basis by the clinical team. Amniotomy was not performed during the study induction period of 24 hours.

Standard procedures

Documentation was similar for all of the studies. The following records were maintained: consent, a comprehensive history and examination, special investigations, medication administration, adverse events, interval to delivery/abortion, details of delivery and estimated blood loss, patient satisfaction visual analogue scores and results of laboratory assays.

Maternal venous blood samples were taken by antecubital venepuncture, prior to administration of medication at 08H00 and 20H00, from the first day of treatment until commencement of prostaglandin (see *Laboratory Methods*). A doctor, blinded to the randomisation, supervised the administration of trial medication (after obtaining the blood samples) from the allocated packet that was kept attached to the Patient Record.

Treatment was considered to have failed if uterine evacuation had not occurred within 24 hours of commencing treatment with prostaglandin. Further management, when required, was determined on an individual basis by the clinical team.

Prior to discharge, assessment of pain, acceptability of the procedure and of overall satisfaction with management (global satisfaction) was assessed by visual analogue scores (VAS) on a scale of 1 to 5. With respect to pain, the woman was asked to assess the severity of pain experienced where 1 represented little or no pain and 5 represented unbearable pain. Acceptability of the procedure referred to the acceptability of the oral medication (with respect to side effects) and the method of pregnancy termination where 1 and 5 represented 'completely unacceptable' and 'completely acceptable' respectively. For global satisfaction, 1 represented 'completely unsatisfied' and 5 'completely satisfied'. The scale was shown to the woman, an explanation given and she indicated the score for each question in turn. The same person in all cases provided guidance for completion of the VAS questions. In some cases the terms required translation through an interpreter.

Consent

Informed consent (see *Appendix*) was obtained in all cases, utilising interpreter skills where required. Counselling was offered within the clinical context by medical staff

and a social worker when appropriate and investigation of intrauterine fetal death and fetal anomaly was conducted as applicable in each individual case.

Approval to undertake the studies was obtained from the Ethics Committee of the Faculty of Health Sciences, University of Cape Town.

Authorisation for the use of trilostane, which is unregistered medication in South Africa, was obtained from the Medicines Control Council (MCC), Department of Health, Republic of South Africa. The use of trilostane was subject to regular review and follow-up reports and was conditional on the reporting of all serious adverse events to the MCC.

Medication

Trilostane (Modrenal® 120 mg, Sanofi-Winthrop, Newcastle-upon-Tyne), an inhibitor of the 3β-hydroxysteroid dehydrogenase enzyme complex which converts pregnenolone to progesterone in the adrenal gland, ovary and placenta, is discussed in detail in the introduction, page 27.

Misoprostol (Cytotec®, Searle, SA), used in Study 1 and Study 2, is a prostaglandin E_1 analogue and is discussed in the introduction, page 30.

Dinoprostone (Prandin® 1 mg – Upjohn, SA) was used in Study 3. This synthetic analogue of prostaglandin E_2 is discussed on page 31.

Oxytocin (Syntocinon®, Alliance, SA) was used in Study 3 and is discussed on page 31.

Randomisation

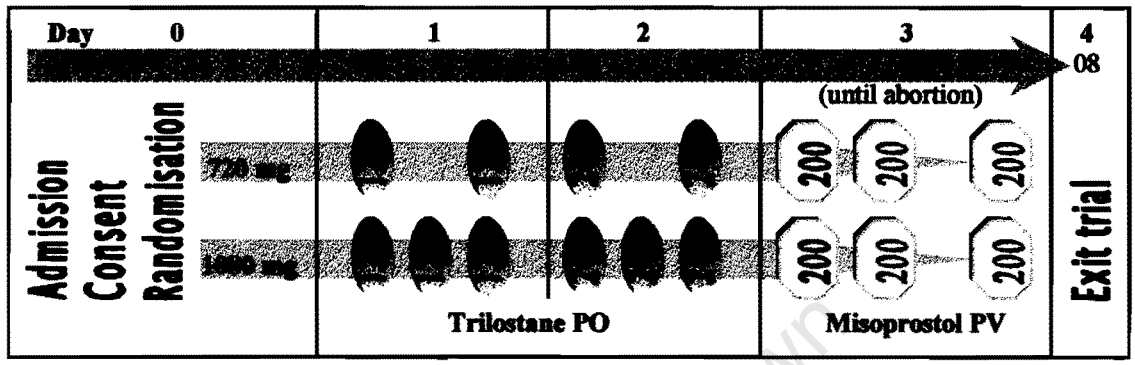
In the placebo-controlled studies, recruited patients were allocated the next in a sequence of numbered packets, randomly containing either trilostane capsules or an

equal number of placebo capsules of identical appearance provided by the manufacturer. Randomisation was carried out by members of the study group who had no direct involvement in recruitment or management of trial patients or in data collation or assessment. Randomisation was performed in blocks of four and the sequence was generated using a random number table. Patients, clinical staff and investigators were blinded to the allocation.

The midtrimester TOP trilostane dose-comparison study was not blinded. Recruited patients were allocated the next in a numbered sequence of packets randomly containing either six or nine trilostane capsules (720 mg or 1080 mg respectively). Randomisation was performed in the same way as for the placebo-controlled studies.

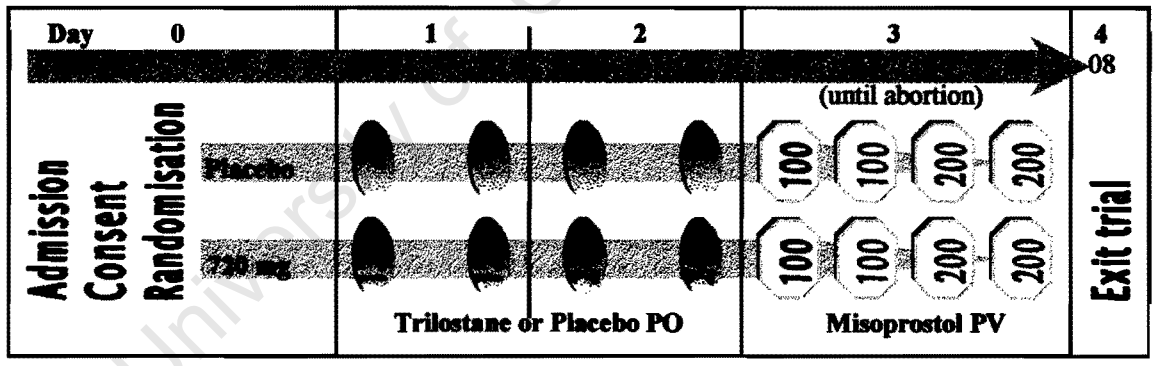
Diagrammatic representation of three clinical trials assessing the efficacy of trilostane in reducing the interval to delivery after prostaglandin administration

Study of midtrimester termination of pregnancy with misoprostol following pre-treatment with trilostane in one of two dose regimens



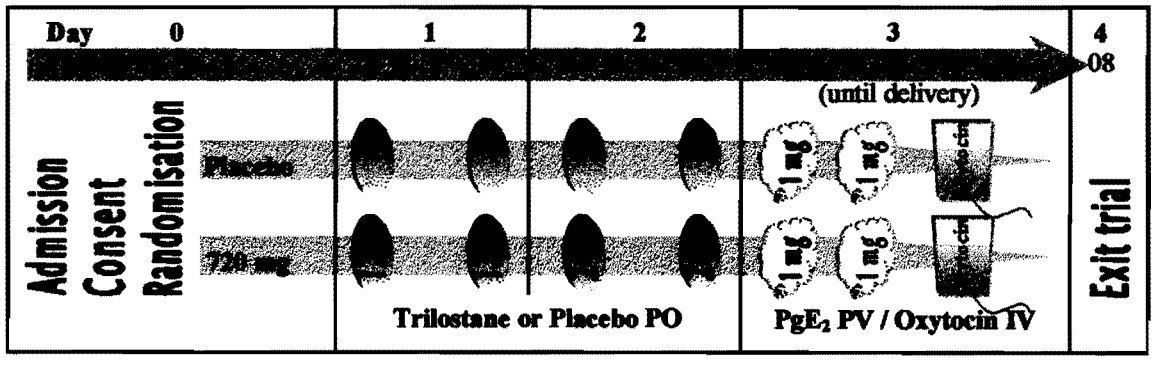
Exit trial = transfer to the clinical team

Placebo-controlled study of termination of pregnancy, for fetal anomaly, with misoprostol following pre-treatment with trilostane



Exit trial = transfer to the clinical team

Placebo-controlled study of induction of labour, for intrauterine fetal demise, with prostaglandin E2 and oxytocin following pre-treatment with trilostane



Exit trial = transfer to the clinical team

Power calculations

Trial size was dictated by a limited licence for the use of trilostane in South Africa. This was a condition of the consent from the Medicines Control Council of South Africa. P*ost-hoc* calculations are possible and will be addressed in the discussion of trial results.

Statistical Analysis

Normally distributed variables are presented as means with either the 95% confidence interval (CI) of the mean or standard deviation. Where log-transformation was used to achieve normality, such as the induction-to-abortion interval, geometric means and 95% CI of the means is reported. Comparisons of continuous, normally distributed variables were conducted using analysis of variance (ANOVA), reporting the 95% CI for the difference or the ratio of the means for normal and log-transformed variables respectively. Non-normally distributed variables were compared using the Mann-Whitney *U* test, Kruskal-Wallis ANOVA or Median test as appropriate. Fisher's exact test was used to compare the incidence variables between the groups. Correlation was assessed by the Spearman rank coefficient.

Laboratory Methods

Venous blood samples were obtained by antecubital venepuncture. This was performed by an attending doctor prior to the administration of trial medication at 08H00 and 20H00 each day. The first sample was taken prior to the first dose of trial medication (08H00) and four further samples were obtained, culminating at 08H00 prior to prostaglandin administration.

Each sample was collected into three 10ml vacutainer tubes (sterile interior) containing gel and clot inactivator. The tubes were individually labelled with patient name, date and time. They were stored upright at 4 °C until centrifuge (within 48 hours of collection). Centrifuge was performed at 5000 rpm for 10 minutes, or until separation was completed, and serum decanted into labelled (trial number, collection date and time) Cryo.s® 5ml screw cap tubes (Greiner Labortechnik) that were then stored at –20 °C until assay.

Serum progesterone and cortisol assays were performed on all samples. Serum estriol and human placental lactogen assays were performed in Study 3, on the pre-treatment samples only. All assays were performed in the Combined Endocrine Laboratory of the Departments of Obstetrics and Gynaecology, Medicine and Chemical Pathology of the University of Cape Town utilising commercially available kits. All samples for an individual patient were measured in the same assay.

Internal and external quality control is exercised for all assays in the laboratory. Statistical methodology applied to these assays involved the following.

1) An assessment of the coefficient of variation (%CV) of the dose response (standard) curve and parameters including the binding, non-specific binding, and displacement in terms of ED20, ED50, and ED80. Acceptable drifts for these

parameters were obtained by assessment of inter-assay variations, of between 5 and 10 assays in the laboratory, and calculating limits for each parameter based on its mean ± 2 standard deviations. An assay was rejected if any two parameters failed the required limits.

2) Three control samples, 'low', 'medium' and 'high' concentration, were measured in duplicate in every assay. An assay was rejected if more than one of these controls were beyond prescribed limits. When only one control was out of range, random samples that had fallen within the 'aberrant' range of that assay were re-assayed.

Progesterone assay

Progesterone was measured by radioimmunoassay (RIA), using a commercially available kit, 'Coat-a-Count', from Diagnostic Products Corporation, catalogue number TKPG.

The kit utilises a solid-phase radioimmunoassay procedure using ^{125}I-labelled progesterone as the tracer. An immobilised antibody-coated tube enables separation of the antibody-bound fraction (of progesterone) by decanting the supernatant. Using this kit, progesterone is measured directly in the serum or plasma and no extraction procedure is required in the assay.

Because of the anticipated high progesterone concentrations in pregnancy, samples were diluted 1:10 in assay diluent prior to assay.

Recovery from 'spiked' samples over the range of the assay was 96 to 100%.

Sensitivity of the assay for the dose response curve was 0.2 nmol/l. Sample sensitivity was 1.2 nmol/l.

Specificity: the antiserum displays the following cross-reactivity.

Progesterone	100%	17α-hydroxyprogesterone	0.3%
Androstenedione	ND	medroxyprogesterone	ND
Corticosterone	0.4%	pregnane	ND
cortisol	ND	5β-pregnan-3α-ol-20-one	0.2%
danazol	ND	5α-pregnan-3,20-dione	0.8%
11-deoxycorticosterone	1.7%	5β-pregnan-3,20-dione	1.3%
11-deoxycortisol	2.4%	pregnenolone	ND
20α-dihydroprogesterone	2.0%	testosterone	ND
estradiol	ND		

ND – not detectable

The coefficient of variation (%CV) for *assay duplication* as a mean of all operators was 4.6%.

The *within-assay* (intra-assay) %CV for levels over the range of the assay was 7.4% and the *between-assay* (inter-assay) %CV was 9.1%.

Cortisol assay

Serum cortisol was measured on the Chiron Diagnostics ACS:180 automated chemiluminescent immunoassay system from Ciba Corning. The assay is a competitive immunoassay using direct chemiluminescent technology and is based on immuno-chemiluminescent assay (ICMA) principles and techniques.

Sample *sensitivity* of the assay was 5.5 mIU/ml.

Specificity: the following cross-reactivity is reported for the assay.

Endogenous Steroids		Synthetic Steroids	
Deoxycorticosterone	0.5%	prednisolone (100 μg/dl)	27.0%
11-deoxycorticosterone	0.4%	6-methyl-prednisolone	20.9%
11-deoxycortisol	7.3%	Dexamethasone	0.2%
21-deoxycortisol	4.5%	Prednisone	6.6%
11-β-hydroprogesterone	0.5%		
17-β-hydroprogesterone	0.5%		

ND – not detectable

The *within-assay* (intra-assay) coefficient of variation (%CV) for levels over the range of the assay was 6.1% and the *between-assay* (inter-assay) %CV was 8.4%.

Estriol assay

Free estriol (FreeE3) was measured by radioimmunoassay using a commercially available kit, 'Coat-A-Count Free Estriol', from Diagnostic Products Corporation (DPC), catalogue number TKEF1.

The kit utilises a solid-phase radioimmunoassay procedure using ^{125}I-labelled estriol as the tracer. An immobilised antibody-coated tube enables separation of the antibody-bound fraction (of estriol) by decanting the supernatant. Using this kit, estriol is measured directly in the serum or plasma and no extraction procedure is required in the assay.

The *reportable range* of the assay was 0.25 to 30.0 ng/ml.

Sample *sensitivity* of the assay was 0.04 ng/ml.

Specificity: the following cross-reactivity is reported for the assay.

Estriol-3-sulphate	0.46%	Oestrone-3-sulphate	ND
Estriol-3β-glucoronide	0.26%	16-Epiestriol	0.26%
Estriol-16β-glucoronide	0.66%	17-Epiestriol	0.10%
Estriol-17β-glucoronide	ND	Cortisol	ND
Estradiol	0.13%	11-Deoxycortisol	ND
Estrone	0.05%	5a-Dihydrotestosterone	ND
Estrone-β-glucoronide	ND	Testosterone	0.003%

ND – not detectable

No assay protein effect was found at protein concentrations of 3.5-14 g/dl.

The *within-assay* (intra-assay) %CV for levels over the range of the assay was 17.1% for values < 0.74 ng/ml and 4.7% for values >0.74 ng/ml.

The *between-assay* (inter-assay) %CV was 21.2% for values <2.9 ng/ml and 9.9% for values, over the assay range, >2.9 ng/ml.

Human Placental Lactogen assay

Human placental lactogen (hPL) was measured by radioimmunoassay using a commercially available kit, 'Coat-A-Count HPL', from Diagnostic Products Corporation (DPC), catalogue number TKHP1.

The kit utilises a solid-phase radioimmunoassay procedure using ^{125}I-labelled hPL as the tracer. An immobilised antibody-coated tube enables separation of the antibody-bound fraction (of estriol) by decanting the supernatant. Using this kit, hPL is measured directly in the serum or plasma and no extraction procedure is required in the assay.

The assay was performed with the 'Basic 25ul sample volume Procedure' as recommended by the kit manufacturer. This protocol provides a *reportable range* of 0.12 to 15.0 μg/ml

Sample *sensitivity* of the assay was 0.12 μg/ml.

Specificity: the manufacturer describes the antiserum to be highly specific for hPL and virtually free of protein and other matrix effects. The following cross-reactivity is reported for the assay.

hCG	ND	LH	ND
FSH	0.04%	Prolactin	0.006%
hGH	0.0003%	TSH	0.012%

ND – not detectable

The *within-assay* (intra-assay) %CV for levels over the range of the assay was 16.0% for values <1.6 µg/ml and 3.6% for values >1.6 µg/ml.

The *between-assay* (inter-assay) %CV was 15.9% for values <1.6 µg/ml and 6.2% for values, over the assay range, >1.6 µg/ml.

RESULTS

Midtrimester termination of pregnancy

Recruitment

Twenty-eight women were recruited to this study and 14 women were randomised to each group. The groups were similar when compared for age (1-way ANOVA), parity (Fisher's exact test) and gestational age (Median test) (Table 2).

Table 2. Characteristics of patients in the two study groups.

	Group 1 (n=14)	Group 2 (n=14)
Age in years – mean (95% CI)	27.5 (23.7-31.3)	27.3 (23.1-31.6)
Number (%) of primigravidae	4 (28.6)	5 (35.7)
Gestational age in weeks – mean (95% CI)	14.29 (13.4-15.1)	15.36 (14.4-16.3)

No significant differences were detected between the groups.

Withdrawals

Two women were withdrawn from the study, one in Group 1 (720 mg trilostane) due to a protocol violation and one in Group 2 (1080 mg trilostane) due to side effects. Details are presented in Table 3.

Table 3. Characteristics of women withdrawn from the trial

Group	Age (years)	Parity	Gestation (weeks)	Reason for withdrawal	Day of withdrawal	IA Interval (hours)
Group 1	38	4	13	Protocol violation.	Day 3	> 72*
Group 2	27	1	15	Side effects	Day 1	< 24§

*No response to misoprostol (additional 400 µg x 6 over 72 hours, orally). Dilatation and evacuation Day 6.

§Delivery after misoprostol 600 µg bolus vaginally.

IA Interval – induction-to-abortion interval

Induction-to-abortion interval

The mean (geometric) induction-to-abortion interval was 9.78 hours (95% CI of 7.52 to 12.68) for Group 1 (n=13) and 8.91 hours (95% CI of 6.47 to 12.28) for Group 2 (n=13). The range was 4.42 to 19.42 hours in Group 1 and 3.30 to 20.10 hours in Group 2. No significant difference was found between the groups (1-way ANOVA and 95% CI of the ratio is 0.74 to 1.62). The cumulative abortion rate over time is shown in Figure 1.

No women aborted or experienced pain or significant bleeding prior to the administration of misoprostol. Of the 26 women included in the analysis, all delivered within 24 hours of commencing misoprostol. The woman withdrawn from Group 1 due to a protocol violation had received the full course of trilostane but the vaginal misoprostol was incorrectly administered (misoprostol tablets were found in her bed). Misoprostol was subsequently administered orally with abortion occurring within 48 hours. The woman withdrawn from Group 2, due to side effects of trilostane

(having received two doses of 120 mg), aborted within 24 hours of misoprostol administration.

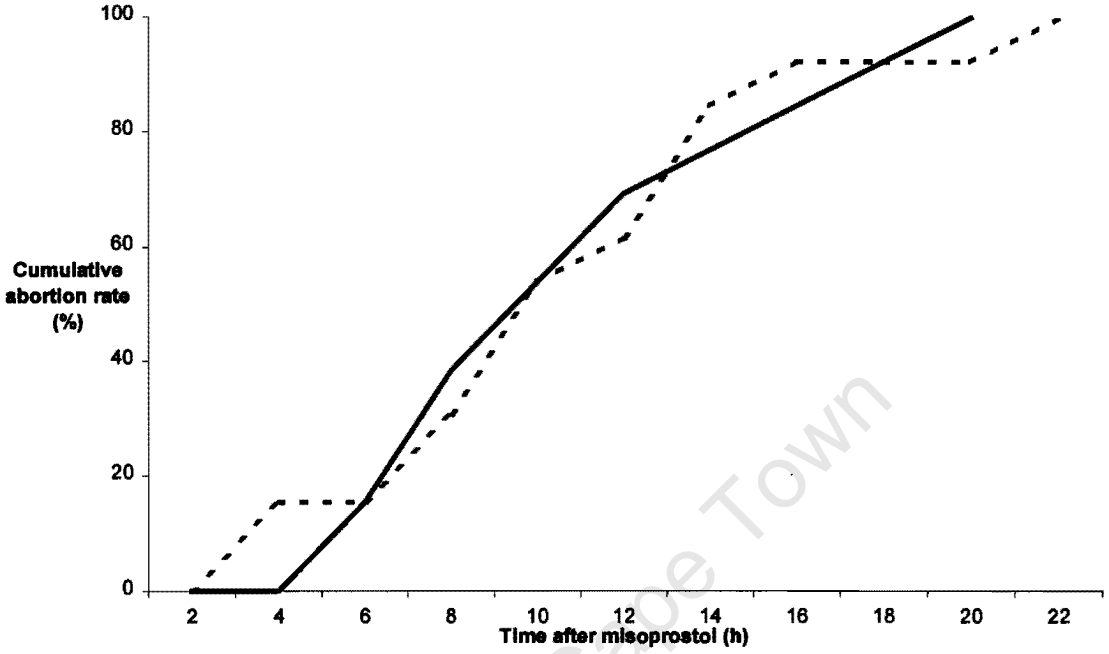


Figure 1. Cumulative abortion rates for midtrimester termination of pregnancy with misoprostol in two groups pre-treated with trilostane. Trilostane was administered in a total dose of 720 mg in Group 1 (—) and 1080 mg in Group 2 (----) respectively.

Side effects, pain and acceptability

Side effects, during trilostane treatment, were reported in 6 women in Group 1 (43%, n=14) and 10 women in Group 2 (71%, n=14). These are summarised in Table 4.

Table 4. Frequency of reported side effects during trilostane administration

Side effects	Group 1 (%) (*n*=14)	Group 2 (%) (*n*=14)
Nausea and/or vomiting	4 (28.6)	7 (50)
Facial itching, burning, oedema	2 (14.3)	5 (35.7)
Headache	3 (21.4)	2 (14.3)
Diarrhoea	—	1 (7.1)
All side effects reported	6 (43)	10 (71)

No significant differences were found between the groups (Fisher's exact test).

Acceptability of the procedure and global satisfaction did not differ significantly between the groups (Kruskal-Wallis ANOVA). The severity of pain was significantly greater in Group 2 (mean VAS 3.23) compared to Group 1 (mean VAS 1.92), *P*=0.005 (Kruskal-Wallis ANOVA). A summary is presented in Figure 2.

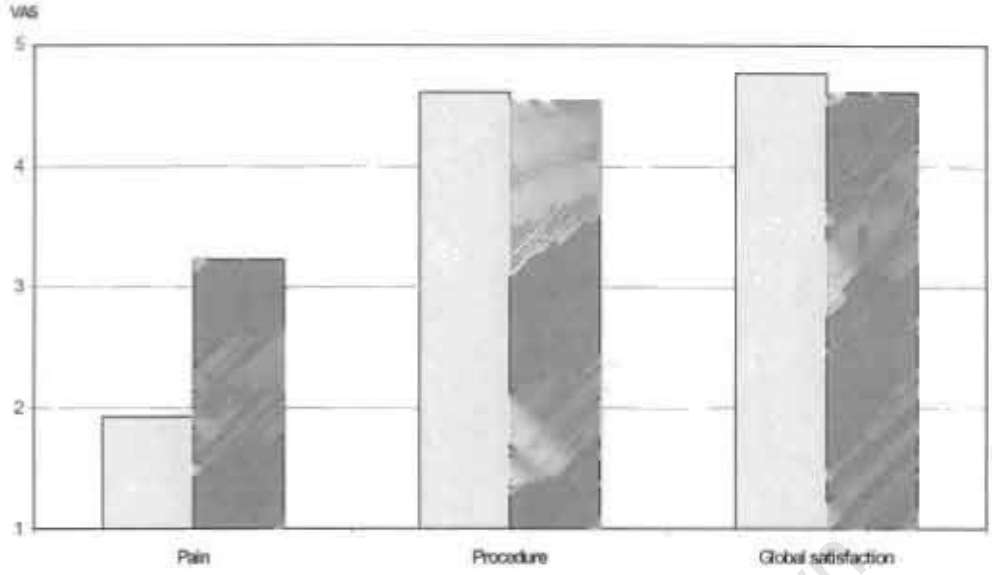


Figure 2. Comparison of pain, acceptability of the procedure and global satisfaction in two groups after midtrimester termination of pregnancy with misoprostol following pre-treatment with trilostane. Visual analogue scores (VAS) are expressed as means for clarity. Group 1 (light grey) and Group 2 (dark grey) received 720 mg and 1080 mg of trilostane respectively. The difference in pain scores was significant (P=0.005, Kruskal-Wallis ANOVA). There was no significant difference in acceptability of the procedure or global satisfaction. Pain: 1= none/minimal, 5= unbearable. Procedure: 1= completely unacceptable, 5= completely acceptable. Global satisfaction: 1= completely unsatisfied, 5= completely satisfied.

Steroidogenesis

Pre-treatment serum progesterone levels were similar between the groups (1-way ANOVA). Comparing pre- and post-treatment progesterone levels, a significant fall from baseline was clearly demonstrated in both groups ($P<0.0001$, one-tailed paired t-test). This is illustrated in Figure 3 where mean progesterone concentrations are expressed as a percentage of pre-treatment levels for clarity.

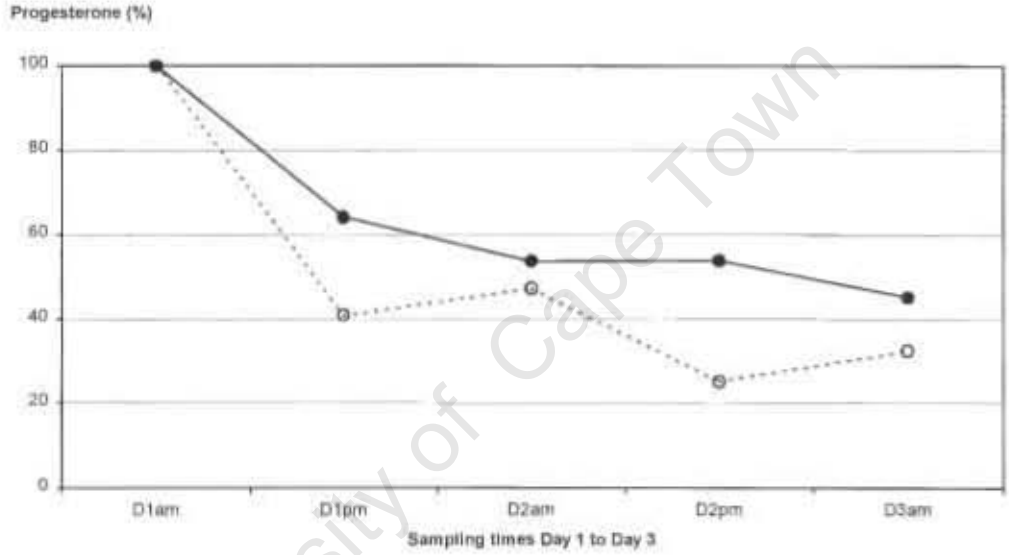


Figure 3. Percentage fall in mean serum progesterone levels in two groups during treatment with trilostane prior to midtrimester termination of pregnancy with misoprostol. Group 1 (—) and Group 2 (----) received 720 mg and 1080 mg of trilostane respectively. The percentage fall in progesterone concentrations over the treatment period (D1am to D3am) was significantly greater in Group 2 than in Group 1, $P= 0.007$.

The effect of trilostane on serum progesterone, assessed by comparison of post-treatment levels (as percentages of pre-treatment levels), was significantly greater for Group 2 (median 31.5%, range 21.9 to 46.9) than for Group 1 (median 42.53%, range 29.6 to 90.1), P=0.007 (Kruskal-Wallis ANOVA, 95% CI of 3.42 to 20.25). This contrasts with a similar magnitude of progesterone suppression when comparing the higher dose regimens (Figure 4).

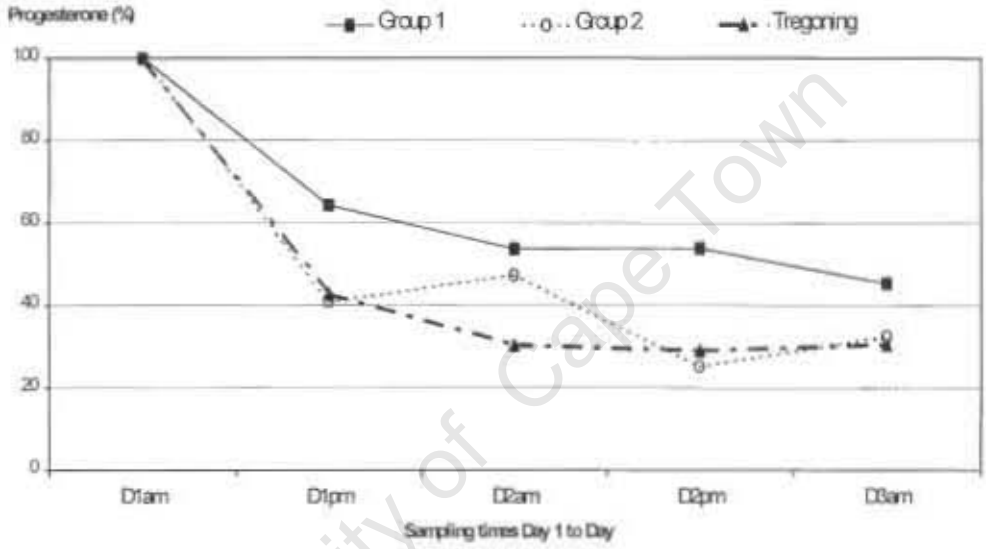


Figure 4. The influence of trilostane dose on mean progesterone levels (as a percentage of pre-treatment levels) in women during treatment with trilostane over 48 hours prior to midtrimester termination of pregnancy with misoprostol. The dosage groups illustrated are: Study 1 Group 1 (720 mg), Study 1 Group 2 (1080 mg) and the trilostane group (1440-2400 mg) from a placebo-controlled study by Tregoning *et al.*[28]

Changes in progesterone levels relative to induction-to-abortion intervals are summarised in Table 5.

Table 5. Induction-to-abortion intervals with serum progesterone levels at 24 and 48 hours of trilostane treatment. Serum progesterone concentrations are expressed as a percentage of pre-treatment levels.

Group 1 (720 mg)			Group 2 (1080 mg)		
IA Interval (hours)	*24 hours (%)*	*48 hours (%)*	*IA Interval (hours)*	*24 hours (%)*	*48 hours (%)*
4.42	47.13	43.68	3.30	24.73	42.86
6.00	46.26	49.66	3.50	44.52	43.49
6.75	85.57	59.70	6.30	46.86	36.82
7.50	58.70	39.13	6.75	33.06	57.66
7.75	48.92	41.94	8.50	24.30	38.32
8.50	59.25	35.27	9.00	38.52	72.79
9.67	57.04	90.14	9.67	31.46	78.09
11.00	55.50	66.06	10.70	21.91	37.64
12.00	55.95	35.24	12.25	31.16	46.38
13.50	40.79	29.61	12.30	28.44	48.00
15.42	51.66	43.13	12.50	24.57	32.08
16.17	54.62	34.87	15.25	43.26	59.55
19.42	41.06	33.33	20.10	33.33	44.23

IA Interval – induction-to-abortion interval

Normal diurnal variability of cortisol was maintained in both groups (Figure 5). There was no significant difference between the groups (Kruskal-Wallis ANOVA).

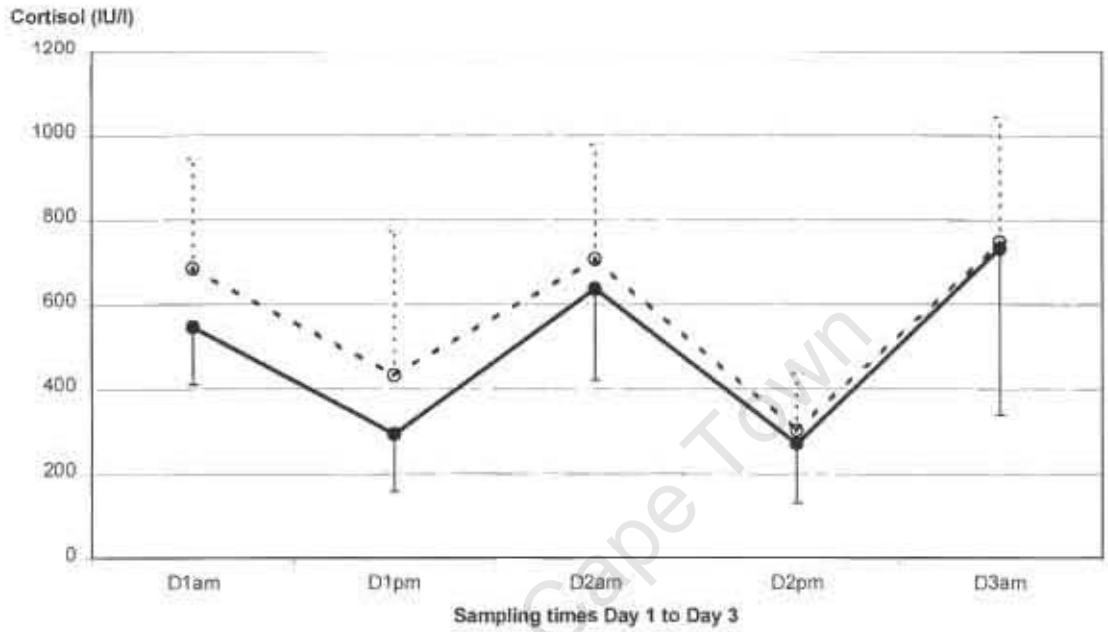


Figure 5. Mean serum cortisol levels with standard deviations in two groups during treatment with trilostane prior to midtrimester termination of pregnancy with misoprostol. Group 1 (—) and Group 2 (----) received 720 mg and 1080 mg of trilostane respectively.

Termination of pregnancy for fetal anomaly

Recruitment

Eleven women were recruited to this study, 6 in the placebo group and 5 in the trilostane group. The groups were similar when compared for age (Median test), parity (Fisher's exact test) and gestational age (Median test) (Table 6).

Table 6. Characteristics of patients in the two study groups.

	Placebo (n = 6)	Trilostane (n = 5)
Age in years – mean (95% CI)	23.5 (20-41)	23 (19-42)
Number (%) of primigravidae	3 (50)	3 (60)
Gestational age in weeks – mean (95% CI)	24 (19-26)	23 (19-30)

No significant differences were detected between the groups

Withdrawals

One woman (in the placebo group) was withdrawn due to a protocol violation; i.e. misoprostol was administered in a reduced dosage regimen (Table 7).

Table 7. Characteristics of woman withdrawn (excluded from primary outcome analysis).

Group	Age (years)	Parity	Gestation (weeks)	Reason for withdrawal	Day of withdrawal	ID Interval (hours)
Placebo	41	5	24	Protocol violation	Day 3	59

ID Interval – induction-to-delivery interval

Induction-to-delivery interval

Due to small numbers, a comparison between the two groups could not be made. Data is summarised Table 8.

Table 8. Summary of women undergoing termination of pregnancy for fetal anomaly, receiving either placebo or trilostane prior to misoprostol.

Trial No	Group	Parity	Gestation (weeks)	Placenta	ID Interval (hours)
1	Placebo	2	20	Complete	11.25
2	Placebo	0	19	Complete	10.75
3	Trilostane	2	30	Complete	63.83
4	Trilostane	0	24	Complete	49.58
5	Placebo	0	26	Complete	17.58
6	Placebo	0	24	Complete	10.33
7	Trilostane	0	19	Complete	16.17
8	Trilostane	0	22	Complete	19.08
9	Trilostane	3	23	Complete	32.58
10*	Placebo	5	24	—	—
11	Placebo	1	24	Incomplete	18.33

ID Interval – Induction-to-delivery interval, *Withdrawn - protocol violation.

Side effects, pain and acceptability

One woman in the placebo group (17%, *n*=6) and four women in the trilostane group (80%, *n*=5) reported side effects. These are summarised in Table 9.

Table 9. Frequency of reported side effects during trilostane administration.

Side effects	Placebo (n = 6)	Trilostane (n = 5)
Nausea and/or vomiting	—	2 (40%)
Facial itching, burning, oedema	—	3 (60%)
Headache	—	1 (20%)
Rash	1 (17%)	—
All side effects reported	1	6

There was no significant difference between the groups (Kruskal-Wallis ANOVA).

There was no significant difference in mean visual analogue scores of pain, acceptability of the procedure or global satisfaction between the two groups. Most women in both groups reported moderate to severe degrees of pain and felt the procedure was more unacceptable than acceptable. Nevertheless, the level of overall satisfaction expressed was more frequently "satisfied" than "unsatisfied" (Figure 6).

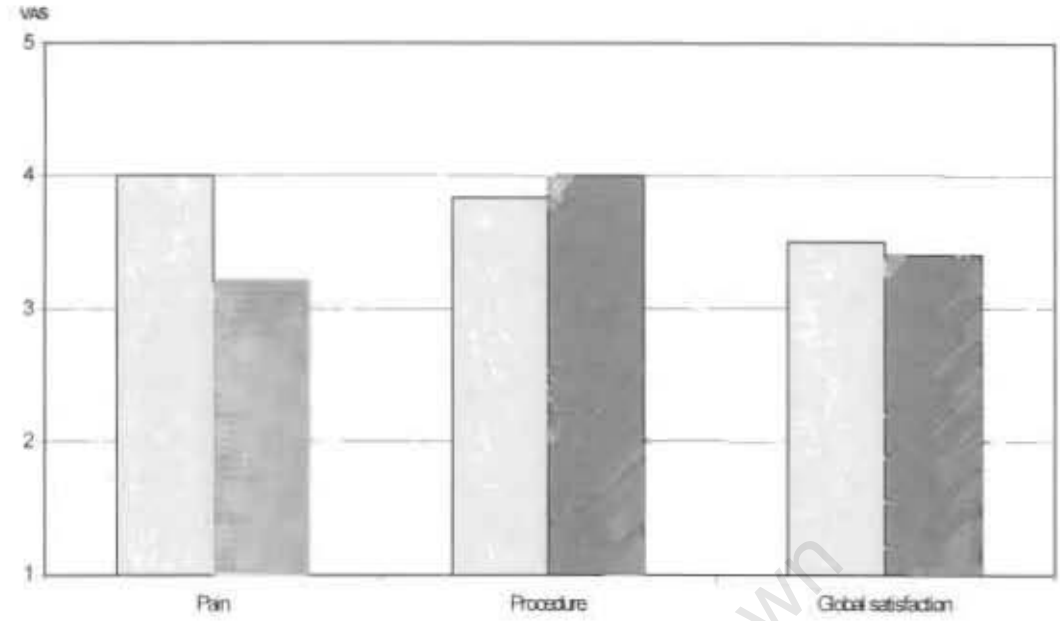


Figure 6. Comparison of pain, acceptability of the procedure and global satisfaction after delivery in two groups that received either placebo (light grey) or trilostane (dark grey) prior to termination of pregnancy for fetal anomaly with misoprostol. Visual analogue scores (VAS) are expressed as means for clarity. There were no significant differences between the groups. Pain: 1=none/minimal, 5=unbearable. Procedure: 1= completely unacceptable, 5= completely acceptable. Global satisfaction: 1= completely unsatisfied, 5= completely satisfied.

Steroidogenesis

Progesterone levels fell significantly for each of the five women receiving trilostane. Mean progesterone levels fell to 57% (significant P=0.01, Paired t-test) and 98% (not significant) of pre-treatment levels in the trilostane and placebo groups respectively. This difference between the two groups was significant (P=0.036, Kruskal-Wallis ANOVA) and is illustrated in Figure 7. Mean progesterone levels were not significantly different between the groups prior to treatment (Kruskal-Wallis ANOVA).

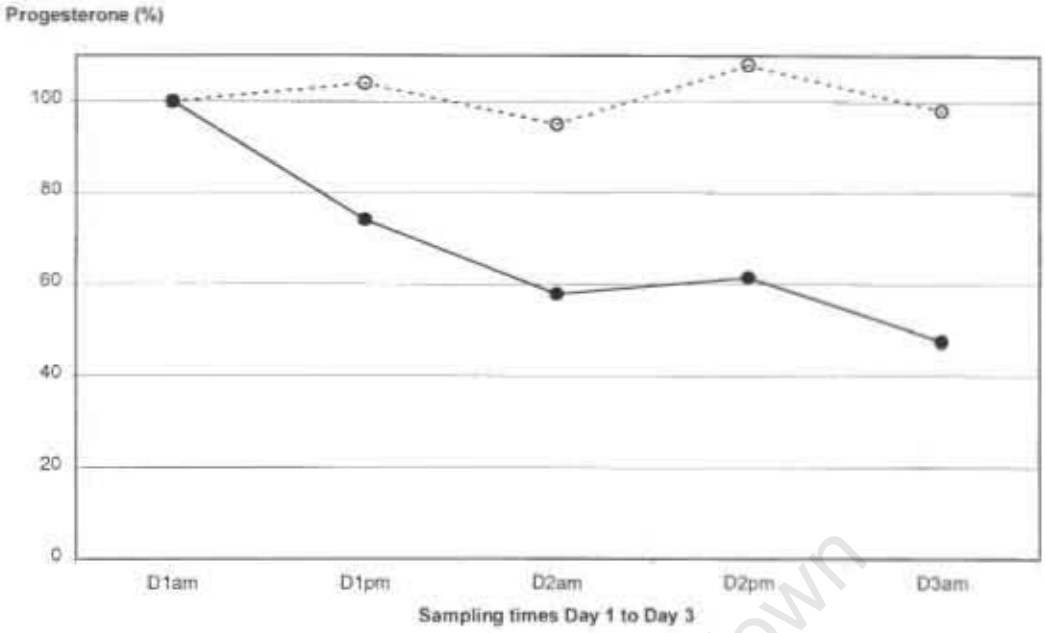


Figure 7. Fall in mean serum progesterone, expressed as a percentage of pre-treatment levels, in two groups during treatment with either trilostane (—) or placebo (----) prior to termination of pregnancy with misoprostol. The percentage fall in progesterone levels in the trilostane group was significantly greater than in the placebo group, P=0.036.

The trilostane regimen in this study was similar to that used in Group 1 of Study 1. In comparison with the earlier gestations treated in Study 1, however, the rate of fall of progesterone was more gradual and of a lower magnitude. This is shown later in Figure 15 in which the mean progesterone concentrations, expressed as a percentage of pre-treatment levels, of the trilostane groups of Studies 1, 2 and 3 are illustrated on one chart for comparison.

Normal diurnal variability of cortisol was maintained in both groups (Figure 8). There was no significant difference between the groups (Kruskal-Wallis ANOVA).

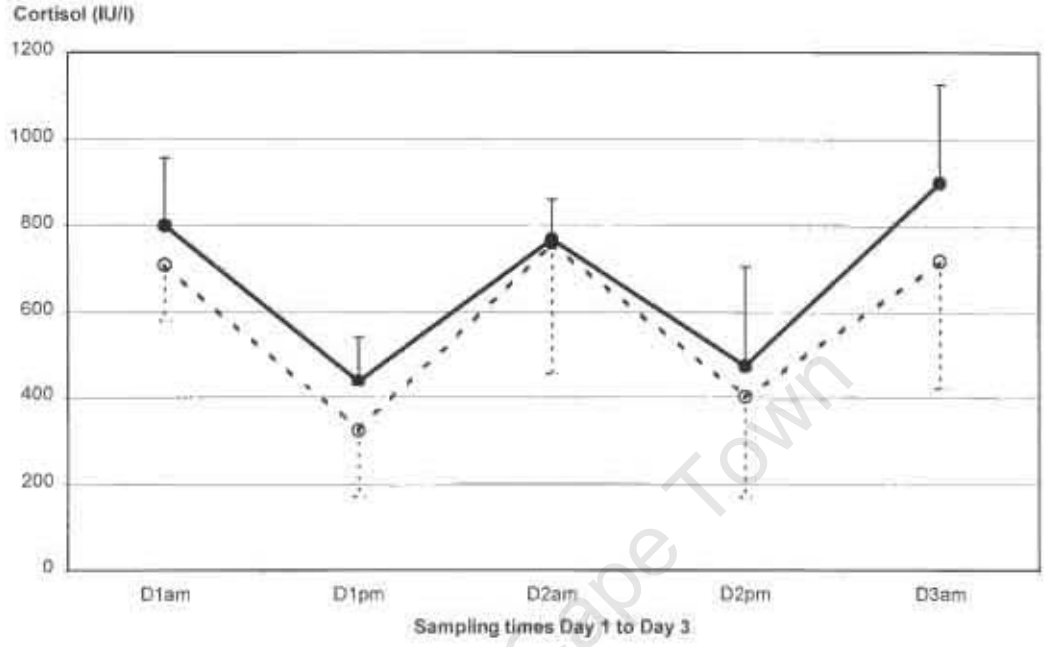


Figure 8. Mean serum cortisol levels with standard deviations in two groups during treatment with either trilostane (—) or placebo (----) prior to termination of pregnancy for fetal anomaly. There was no significant difference between the groups.

Induction of labour for intrauterine fetal death

Recruitment

Twenty women were recruited to this study, 10 in the placebo group and 10 in the trilostane group. The groups were similar when compared for age (1-way ANOVA), parity (Fisher's exact test), gestational age (Kruskal-Wallis ANOVA) and Bishop's score (Median test), summarised in Table 10.

Table 10. Characteristics of patients in the two study groups.

	Placebo (n = 10)	Trilostane (n = 10)
Age in years – mean (95% CI)	28.1 (24.6-34)	29.5 (23.2-33)
Number (%) of primigravidae	4 (40)	3 (30)
Gestational age in weeks – mean (95% CI)	31.0 (28-34)	31.9 (29-35)
Bishop score – median (range)	4 (0-7)	3.75 (0-6)

No significant differences were detected between the groups

Withdrawals

One woman (who received placebo) withdrew consent to participate in the trial after the first dose of trial drug on Day 1 of treatment. This was on the basis of not being prepared to wait two days for induction. She subsequently laboured and delivered spontaneously on Day 2 without additional treatment. As a member of the placebo group, her data was included in the analysis (with her consent).

One woman in the trilostane group delivered on the day of randomisation (Day 0), prior to treatment, and was thus excluded from analysis of treatment efficacy and acceptability.

These details are summarised in Table 11.

Table 11. Characteristics of women withdrawn / excluded from analysis.

Group	Age (years)	Parity	Gestation (weeks)	Reason for withdrawal	Day of withdrawal	TD Interval (hours)
Placebo	22	0	32	Consent withdrawn	Day 1	26.67
Trilostane	27	1	36	Delivery pre-treatment	Day 0	—

TD Interval – Treatment to delivery interval

Induction-to-delivery interval

6 women delivered before prostaglandin administration, 4 in the trilostane group and 2 in the placebo group. The median induction-to-delivery interval in the remaining women was 22.84 hours (95% CI of 8.00 to 40.00) and 15.67 hours (95% CI of 5.70 to 61.12) in the trilostane (n=6) and placebo (n=8) groups respectively. This difference was not significant (Kruskal-Wallis ANOVA).

Treatment to delivery interval

The treatment to delivery interval is defined as the interval from commencement of trilostane or placebo to delivery. The median treatment to delivery interval was 62.00 hours (95% CI of 23.67 to 81.5) for the trilostane group (n=9) and 62.25 hours

(95% CI of 26.67 to 83.8) for the placebo group (n=10). No significant difference was found between the groups (Kruskal-Wallis ANOVA).

The cumulative delivery rate over time is shown in Figure 9 and all results are summarised in Table 12 and Table 13.

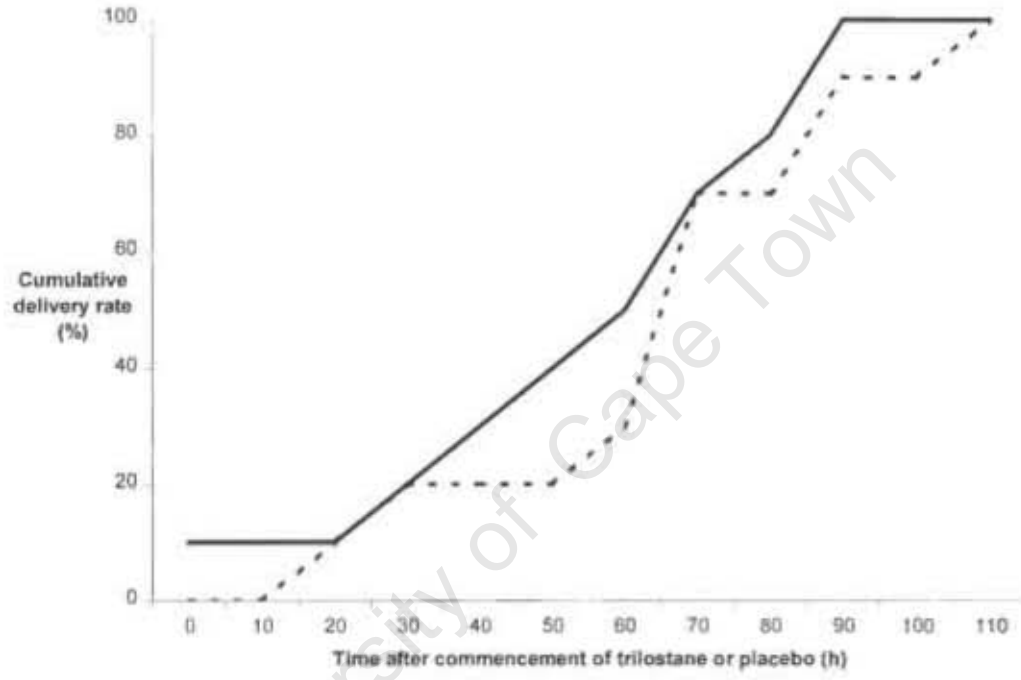


Figure 9. Cumulative delivery rates in 20 women with intrauterine fetal demise treated with either placebo (---- n=10) or trilostane (— n=10) for 48 hours prior to induction of labour with prostaglandin E_2 and oxytocin. One women in the trilostane group delivered prior to commencing treatment.

Table 12. Summary of results of Study 3 - Trilostane group.

Trial No.	Parity	GA (weeks)	IUD* (weeks)	Bishop Score	IDI§ (hours)	TDI† (hours)	Day	Progesterone (nmol/l)	
								AM	PM
2	6	26	<1	2	33.5	81.5	1	244	141
							2	70	111
							3	70	180
3	0	30	<1	1	40.0	88.0	1	249	81
							2	64	70
							3	57	112
6	5	38	<1	6	19.7	67.7	1	598	766
							2	521	442
							3	361	639
7	1	36	<1	4	—	—			
	Delivery before trilostane								
11	0	33	>2	5	—	38.1	1	456	273
	Delivery Day 2: after 480mg of trilostane						2	142	241
12	0	33	?	4	14.0	62.0	1	1004	621
							2	372	223
							3	174	
13	0	26	?	0	26.0	74.0	1	240	164
							2	176	131
							3	120	
14	3	34	?	6	—	43.4	1	953	660
	Delivery Day 2: after 480mg of trilostane						2	735	488
18	4	35	>2	4	8.0	56.0	1	826	626
							2	576	531
							3	344	
19	1	33	>2	3	—	23.7	1	259	238
	Delivery Day 2: after 240mg of trilostane								

GA–gestational age, IUD–intrauterine death, IDI–induction-to-delivery interval, TDI–treatment to delivery interval

*Duration of IUD estimated from ultrasound or post-mortem findings when possible

§Interval from first dose of prostaglandin to delivery

†Interval from first dose of trilostane or placebo to delivery.

Table 13. Summary of results of Study 3 - Placebo group.

Trial No.	Parity	GA (weeks)	IUD* (weeks)	Bishop Score	IDI§ (hours)	TDI† (hours)	Day	Progesterone (nmol/l) AM	PM
1	0	32	<1	3	—	26.7	1	236	
	Delivery Day 2: withdrew after one placebo dose								
4	0	29	1	2	14.5	62.5	1	219	160
							2	199	191
							3	174	140
5	5	33	?	4	12.5	60.5	1	721	777
							2	867	749
							3	671	
8	0	38	?	7	14.0	62.0	1	616	630
							2	597	579
							3	537	
9	3	38	2	5	5.7	53.7	1	406	486
							2	520	411
							3	476	
10	1	26	?	5	—	12.6	1	30	
	Delivery Day 1: after one placebo dose								
15	2	32	?	4	32.6	80.6	1	242	265
							2	210	306
							3	250	
16	2	26	<1	4	16.8	64.8	1	131	100
							2	107	115
							3	123	
17	0	27	<1	0	35.8	83.8	1	176	180
							2	165	167
							3	175	
20	1	29	<1	0	61.1	109.1	1	618	632
							2	589	688
							3	559	

GA – gestational age, IUD – intrauterine death, IDI – induction-to-delivery interval, TDI – treatment to delivery interval.

*Duration of IUD estimated from ultrasound or post-mortem findings

§Interval from first dose of prostaglandin to delivery

†Interval from first dose of trilostane or placebo to delivery. Side effects, pain and acceptability

Side effects, pain and acceptability

Two women in the placebo group (22%, n=9) and four in the trilostane group (44%, n=9) reported side effects, a difference that did not reach significance. Details are summarised in Table 14.

Table 14. Frequency of reported side effects during trilostane administration

Side effects	Placebo (n = 9)	Trilostane (n = 9)
Nausea and/or vomiting	—	1 (11.1%)
Facial itching, burning, oedema	—	2 (22.2%)
Headache	1 (11.1%)	—
Heartburn	1 (11.1%)	1 (11.1%)
All side effects reported	2 (22.2%)	4 (44.4%)

The difference in incidence of side effects between the groups was not significant (Fisher's exact test).

There was no significant difference in mean visual analogue scores of pain, acceptability of the procedure or global satisfaction between the two groups (Figure 10).

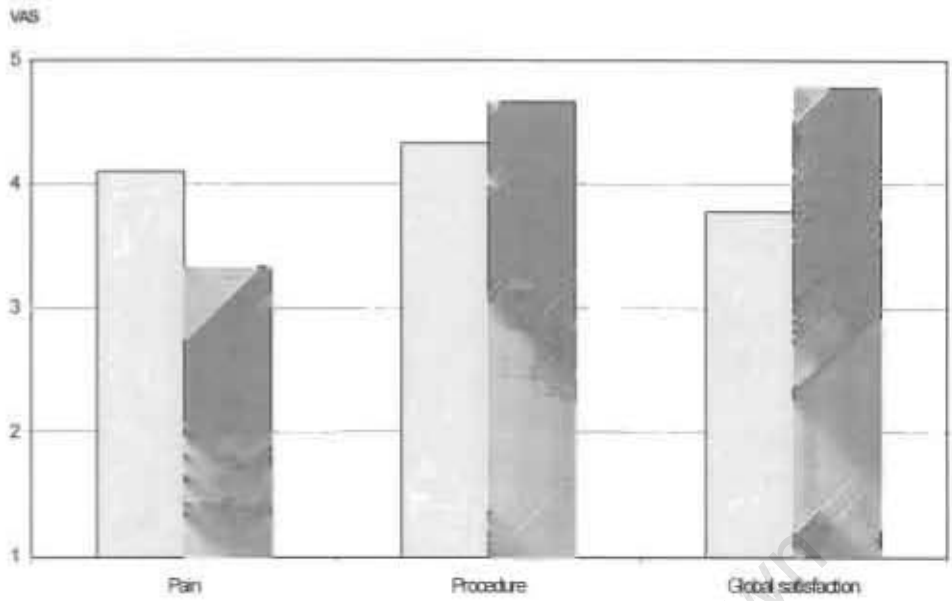


Figure 10. Comparison of pain, acceptability of the procedure and global satisfaction after delivery in two groups that received either placebo (light grey) or trilostane (dark grey) prior to induction of labour for intrauterine death with prostaglandin E_2. Visual analogue scores (VAS) are expressed as means for clarity. There were no significant differences between the groups. Pain: 1=none/minimal, 5=unbearable. Procedure: 1= completely unacceptable, 5= completely acceptable. Global satisfaction: 1= completely unsatisfied, 5= completely satisfied.

Steroidogenesis

Following 48 hours of treatment with either trilostane or placebo, mean progesterone levels were 35% (significant P=0.003, one-tailed paired t-test) and 95% (non-significant P=0.13, one-tailed paired t-test) of pre-treatment levels respectively. The difference between the groups was significant (P=0.001, Kruskal-Wallis ANOVA) and is illustrated in Figure 11 and Figure 12.

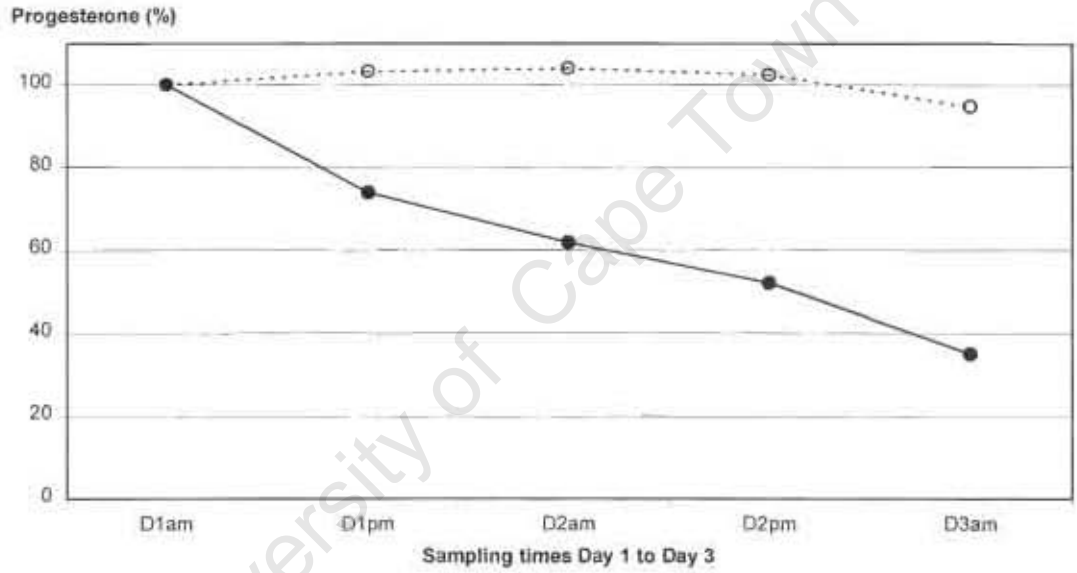


Figure 11. Fall in mean serum progesterone, expressed as a percentage of pre-treatment levels, in two groups during treatment with either trilostane (—) or placebo (----) prior to induction of labour for intrauterine death with prostaglandin E_2. The percentage fall in progesterone levels in the trilostane group was significantly greater than in the placebo group (P=0.001, Kruskal-Wallis ANOVA).

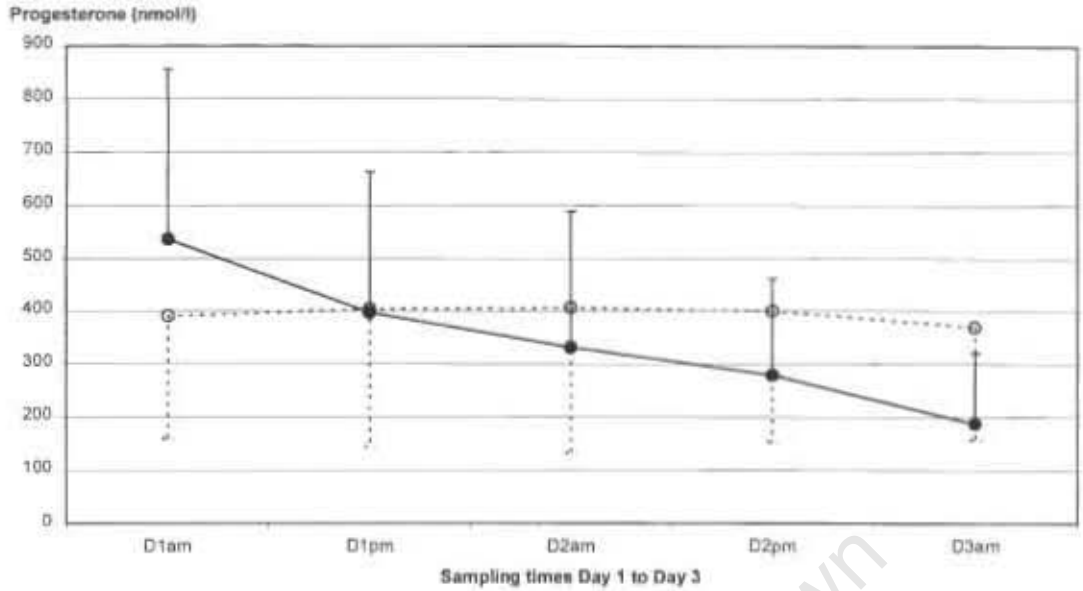


Figure 12. Mean serum progesterone levels with standard deviations in two groups during treatment with either trilostane (—) or placebo (----) prior to induction of labour for intrauterine death with prostaglandin E_2. Levels between the groups did not differ significantly pre-treatment. During treatment, mean progesterone levels remained unchanged in the placebo group and fell significantly in the trilostane group (P=0.003, paired t-test).

No correlation could be demonstrated between either the pre-treatment progesterone levels or the fall in progesterone levels and the intervals to delivery. This data is illustrated in Figure 13 (trilostane group) and Figure 14 (placebo group).

A non-significant positive trend is seen between the pre-treatment progesterone level and the magnitude of fall in serum progesterone over the first 24 hours (ρ_s= 0.69), apparent in Figure 13.

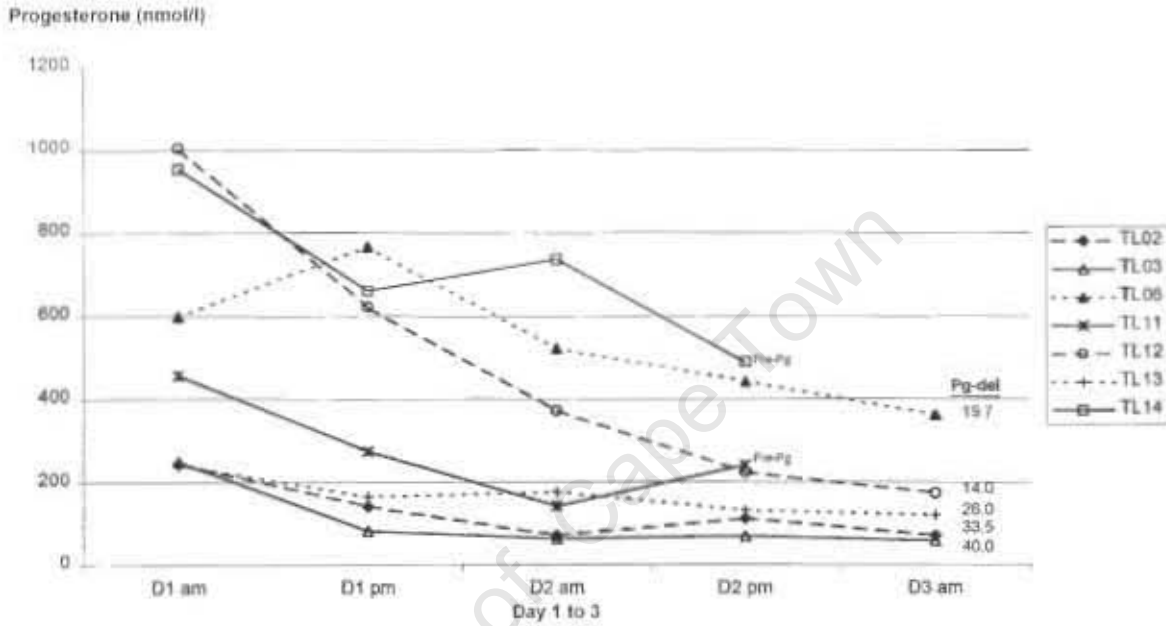


Figure 13. Progesterone levels in women with intrauterine fetal demise during treatment with trilostane for 48 hours prior to induction of labour with prostaglandin.

TL – trial number, Pre-Pg – delivery before prostaglandin induction,

Pg-del – interval from the first dose of prostaglandin to delivery (hours)

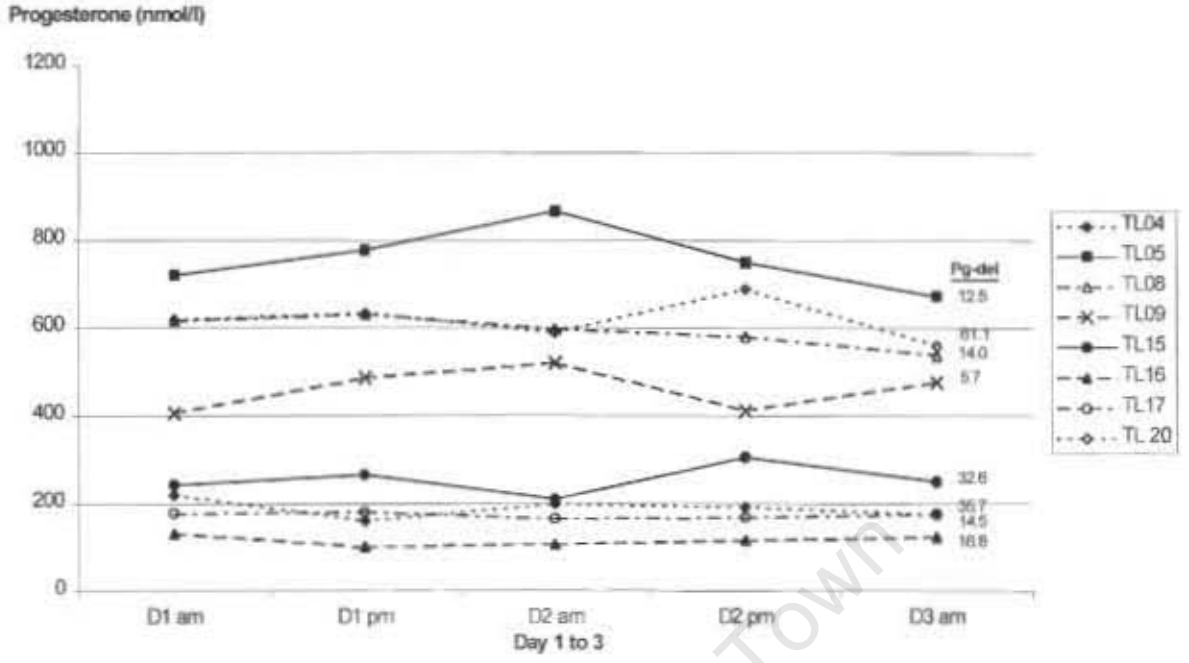


Figure 14. Progesterone levels in women with intrauterine fetal demise during treatment with placebo for 48 hours prior to induction of labour with prostaglandin.

TL – trial number

Pg-del – interval from the first dose of prostaglandin to delivery (hours)

The mean progesterone concentrations (expressed as a percentage of pre-treatment levels), of the trilostane groups in Studies 1, 2 and 3 are illustrated on one chart for comparison (Figure 15).

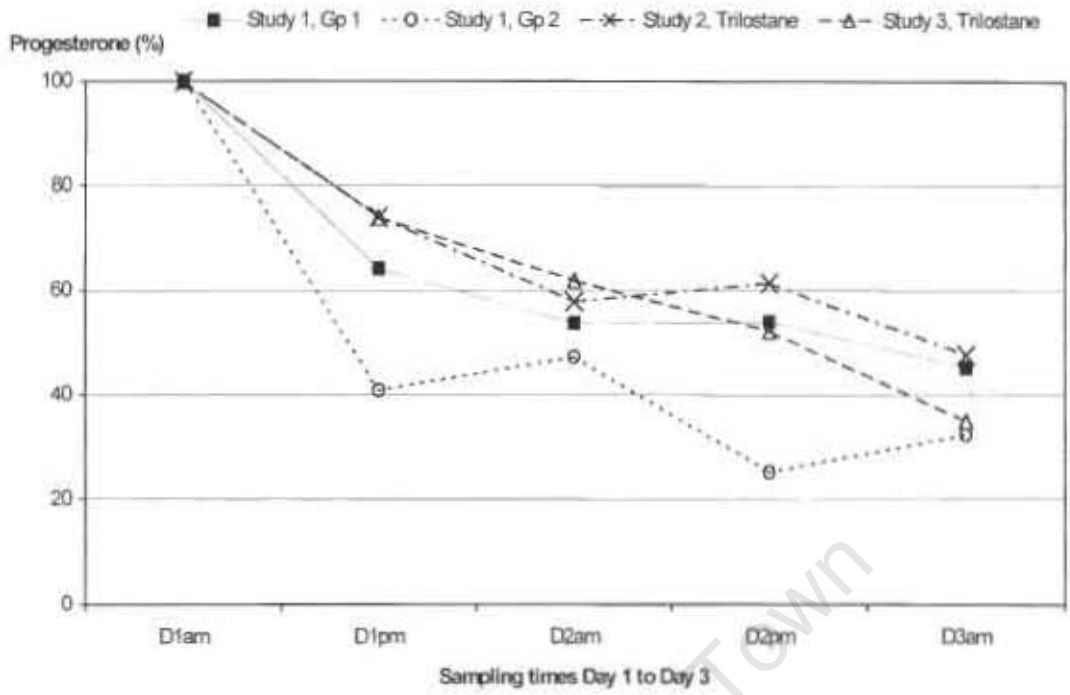


Figure 15. Comparison of mean progesterone levels (as a percentage of pre-treatment levels) in Group 1 (720 mg) and Group 2 (1080 mg) from Study 1 and the trilostane groups in Studies 2 and 3.

Pre-treatment serum progesterone levels correlated positively with pre-treatment serum human placental lactogen (hPL) levels, ρ_s=0.75 (95% CI of 0.46 to 0.90), P=0.0002, as shown in Figure 16. No correlation could be detected between the treatment to delivery or induction-to-delivery intervals and the pre-treatment levels of either serum progesterone or serum hPL.

Pre-treatment levels of serum hPL were within the normal range for gestational age in 6 of 10 (60%) and 7 of 9 (78%) women in the placebo and trilostane groups respectively. Other women had levels below the normal range. This difference was not significant (Fisher's exact test).

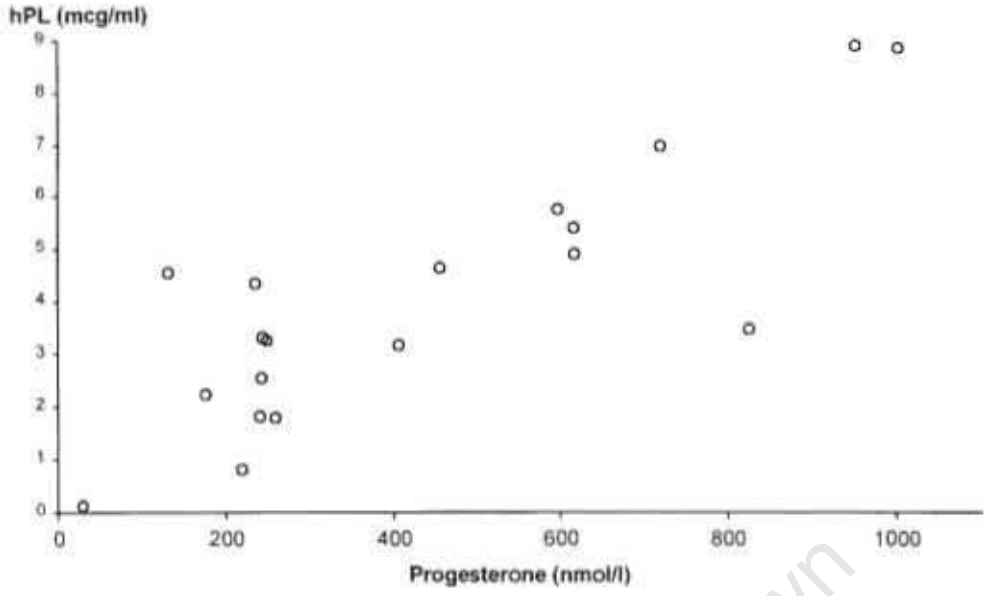


Figure 16. Correlation between pre-treatment serum progesterone and serum human placental lactogen (hPL) levels in 19 women with intrauterine fetal death (ρ_s=0.75, *P*=0.0002).

Pre-treatment serum estriol levels were below the normal range for gestational age in all women and showed no correlation with intervals to delivery or pre-treatment serum progesterone concentrations.

Estriol and hPL results are summarised in Table 15.

Normal diurnal variability of cortisol was maintained in both groups (Figure 17). There was no significant difference between the groups (Kruskal-Wallis ANOVA).

Table 15. Summary of pre-treatment serum levels of hPL and estriol.

Trial No.	Group	GA (weeks)	hPL (µg/ml)	hPL normal range for GA	Estriol (ng/ml)	Estriol normal range for GA	TD Interval
1	P	32	4.34	3.3-10	0.529*	7.5-20	26.67
2	T	30	3.31	2.8-8.2	0.591*	5.2-17.5	81.50
3	T	30	3.25	2.8-8.2	0.239*	5.2-17.5	88.00
4	P	29	0.82*	1.5-7.3	0.240*	5-17.5	62.50
5	P	33	6.98	3.4-10.5	0.083*	7-23	60.50
6	T	38	5.73	3.6-12.6	0.225*	15-40	67.67
7§	T	—	—	—	—	—	—
8	P	38	5.39	3.6-12.6	0.384*	15-40	62.00
9	P	38	3.17*	3.6-12.6	0.365*	15-40	53.70
10	P	28	<0.12*	1.5-7.3	0.136*	5-17.5	12.63
11	T	37	4.64	3.5-12.5	1.477*	10-35	38.08
12	T	33	8.84	3.4-10.5	0.544*	7-23	62.00
13	T	26	1.83	1.5-7.3	0.043*	4.5-15	74.00
14	T	34	8.88	3.4-11.1	0.368*	8-25	43.42
15	P	32	2.53*	3.3-10	0.389*	7.5-20	80.58
16	P	26	4.54	1.5-7.3	0.390*	4.5-15	64.83
17	P	27	2.22	1.5-7.3	1.152*	4.5-15	83.75
18	T	35	3.48*	3.5-12	1.807*	8.5- 27.5	56.00
19	T	33	1.79*	3.4-10.5	0.349*	7-23	23.67
20	P	29	4.89	1.5-7.3	1.326*	5-17.5	109.12

P – placebo, T – trilostane, GA – gestational age, hPL – human placental lactogen, TD Interval – treatment to delivery interval (hours)

*Below normal range, §Delivery Day 0 – no data available

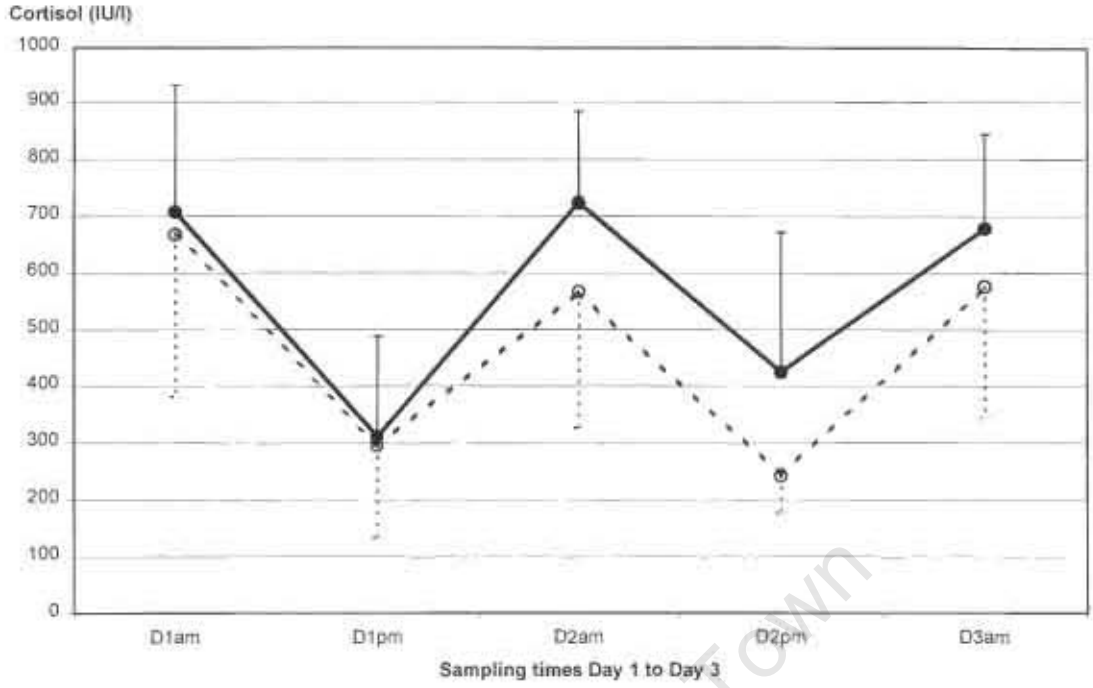


Figure 17. Mean serum cortisol levels with standard deviations in two groups during treatment with either trilostane (—) or placebo (----) prior to induction of labour for intrauterine death with prostaglandin E_2. There was no significant difference between the groups.

DISCUSSION

Study 1 – Mid-trimester Termination of Pregnancy

This study has demonstrated that a low dose of trilostane is effective therapy in second trimester termination of pregnancy with misoprostol. The study design, environment and population group was similar to that of the placebo-controlled trial conducted by Tregoning *et al*,[28] which allows a valid comparison to be made.

Study power

This study had a power of 90% to detect a difference in induction-to-abortion intervals, between Group 1 and Group 2, of 2 hours at a significance level of 5%.[71]

Progesterone inhibition and induction-to-abortion interval

In Tregoning's study of 48 women, the use of trilostane over 48-72 hours (1440-2400 mg) significantly reduced the induction-to-abortion interval with misoprostol when compared to placebo (9.2 hours versus 21.6 hours respectively). Utilising the same misoprostol regimen, the induction-to-abortion intervals in Study 1 of 9.78 hours in Group 1 (720 mg trilostane) and 8.91 hours in Group 2 (1080 mg trilostane) were comparable to Tregoning's trilostane group. This shows that the increase in uterine sensitivity to prostaglandin can be achieved with less than one third of the maximum dose (2400 mg) administered in Tregoning's study. Investigating the required dose of mifepristone in termination of early pregnancy with misoprostol, McKinley and colleagues[72] reported similar efficacy with the lower dose of 200 μg compared to 600 μg. They concluded that this was a consequence of similar peak concentrations of the drug in serum despite the difference in ingested dose. A different explanation is required in the case of trilostane where the magnitude of

progesterone inhibition in (Study 1) Group 1 was significantly less than that seen with Group 2, contrasting with the similarity of suppression between the higher dose regimens (Group 2 and Tregoning's study) shown in Figure 4.

Despite the difference in the magnitude of progesterone suppression between Group 1 and Group 2, the induction-to-abortion intervals were no different between the groups in mean or range. These findings suggest both a maximum suppressive effect on progesterone synthesis as well as a threshold level, significantly higher than the maximally suppressed level, below which uterine sensitivity to prostaglandins increases.

Side effects, pain and adrenal steroidogenesis

Although side effects were reported less frequently in Group 1 compared to Group 2, this difference did not reach significance. Nausea and vomiting and facial symptoms are well know side effects of the medication and are likely to be dose-dependent. In Tregoning's study (personal communication), these side effects occurred with a similar frequency to that of Group 2. It follows that a lower effective dose is likely to be beneficial in terms of a reduced incidence of side effects and any reduction in dose without compromising efficacy will also have cost benefits.

The finding that more pain was reported in Group 2, receiving the higher dose of trilostane is difficult to explain. If this were real (rather than a Type II error), one would expect the higher dose of trilostane used in Tregoning's study to corroborate this with a correspondingly higher pain score. The mean VAS score of 2.23 in his trilostane group is, however, lower than Group 2 in this study.

Trilostane has been registered for use in the United Kingdom since the 1970s (see page 27) but is not licensed for use as an abortifacient. It therefore has a longer term of safety data than mifepristone, which became available for clinical use approximately 10 years later. While both drugs are comparable in safety profiles generally, mifepristone is contra-indicated for use in women at risk of porphyria.[78] Both mifepristone and trilostane have anti-glucocorticoid effects which are not of clinical importance in the doses employed for termination of pregnancy in otherwise healthy individuals. There is no long-term data on teratogenicity with either drug at present. Side effects are a significant problem of both mifepristone and trilostane, the gastro-intestinal complaints being most prevalent. The incidence of nausea and vomiting, 28-50%, in Study 1 is similar to the 36% reported in Tregoning's study[28] (utilising higher doses of trilostane) and compares with the 40-60% incidence reported in mifepristone studies.[27, 74] Unlike mifepristone, some women experience facial symptoms (tingling mouth, itchy eyes, swelling of lips and palate) with trilostane. Theses symptoms are not associated with anaphylaxis or respiratory distress and are usually easily tolerated.

As with mifepristone studies, there was a high degree of acceptability amongst women using trilostane. The single dose regimen for mifepristone, however, contrasts with the requirement for twice-daily administration of trilostane. This has significant implications for medicine control, administration and compliance.

In the United Kingdom, current protocols for second trimester termination of pregnancy recommend a single 200mg tablet of mifepristone (Mifegyne®, Exelgyn) at a cost of approximately £14 (R140) per dose.[78] A single course of trilostane (Modrenal®, Wanskerne), equivalent to 720 mg, would currently cost £6 (R60).[79] Cost considerations are highly significant when establishing a standard protocol in a

health service and the advantages of mifepristone in termination of pregnancy would be eclipsed by cost implications in the developing world.

Study 2 – Termination of pregnancy for fetal anomaly

This study was performed to assess the role of trilostane in pregnancy termination beyond 18 weeks. The need to terminate a pregnancy at this stage most frequently arises when a serious fetal abnormality is detected in the course of antenatal screening and prenatal diagnosis. This is usually achieved by medical induction of labour with or without prior fetocide. In the series reported in this study, fetocide was not used prior to termination and thus an intact fetoplacental unit was present at the time of trilostane therapy.

Study power

Power calculations are difficult due to the lack of sufficiently similar studies (with respect to prostaglandin induction regimens) on which to base expectations. If one uses the induction-to-delivery data from the placebo group in this study as a guideline, the study had sufficient power (90%) to detect a 50% reduction in induction-to-delivery interval at a significance level of 5%.[71] To detect a smaller difference, larger studies would be required. In addition, small numbers are strongly influenced by treatment failures, which were more frequent in the trilostane group.

Prostaglandin dose versus trilostane dose

Although the small numbers limit interpretation of the findings, it is surprising that no trend could be observed when comparing the induction-to-delivery intervals in the trilostane and placebo groups. As noted previously (page 64), the fall in progesterone levels observed in the trilostane group was more gradual and of a lower magnitude in comparison with the earlier gestations of Study 1 (Figure 15). Furthermore, an initial

dose of 100 μg of misoprostol was used for induction in this study in contrast to the 200 μg dose used in Study 1. A lack of data regarding the safe use of misoprostol beyond 20 weeks gestation together with an anticipated augmentation of endogenous prostaglandin levels due to progesterone inhibition were reasons for commencing induction with the lower dose of misoprostol.[12]

Prior to the advent of antiprogestin therapy, Csapo and Pulkinnen proposed the need for a 'prostaglandin impact' in medical termination of pregnancy.[80] This was achieved with an initial large bolus of prostaglandin that would cause uterine contraction as well as disrupting the feto-placental unit leading to a fall in progesterone levels and stimulating endogenous prostaglandin production. Thus, a 'second phase' of endogenous prostaglandin production would be responsible for completing the abortion process. The use of an antiprogestin to reduce progesterone levels prior to the administration of prostaglandin has been suggested as a substitute for such a 'prostaglandin impact' strategy.[81] If the rate of progesterone withdrawal were a factor in the efficacy of antiprogestin therapy then ensuring rapid and adequate progesterone withdrawal at the cellular level in the context of this study would probably require an increase in trilostane dosage. To what degree the duration of progesterone suppression influences efficacy and whether this varies with gestational age is also unclear. Whether a higher dose and/or a longer period of trilostane administration would yield better results requires further investigation. Likewise, the optimal dose of prostaglandin in these circumstances remains to be determined.

Study 3 – Induction of labour for intrauterine death

Induction-to-delivery interval

This study has not been able to demonstrate a beneficial role for the use of trilostane prior to induction of labour with prostaglandin in women with intrauterine fetal demise. Possible reasons include small trial numbers, sample heterogeneity and questionable efficacy of the selected drug regimens.

The aim of the study was to determine whether the use of trilostane prior to prostaglandin induction of labour would result in a shortened prostaglandin to delivery interval. Trilostane was administered for 48 hours usually commencing the day following diagnosis of intrauterine death. Importantly, and not surprisingly, a number of women laboured spontaneously, delivering prior to commencement of prostaglandin. One randomised woman delivered prior to commencing trilostane. This finding is not reported in other studies of active management of intrauterine death where all but one had commenced induction with either prostaglandin or antiprogestin soon after admission.[43] When intrauterine death occurs due to placental abruption, spontaneous labour frequently follows shortly thereafter or labour is easily induced by amniotomy and oxytocin infusion.[82] Lesser degrees of placental separation, with a surge in endogenous prostaglandins, may not be as obvious and will naturally contribute a proportion of early deliveries in all studies. In Study 3, the exclusion of women in whom a diagnosis of placental abruption had been made as well as any for whom a delay of 48 hours would be unwise increases potential differences between the cohort of women in Study 3 and previous studies[32-38, 41, 42, 47-54] and seriously limits comparisons.

The only other study that has assessed the role of progesterone inhibition prior to induction of labour with prostaglandin in the management of intrauterine fetal death was performed by Selinger[43] in 1988, using epostane. He did not report any deliveries prior to induction with PGE_2, during the 24-hour interval for administration of epostane. His study, however, included only two pregnancies beyond 24 weeks gestation and was not prospectively controlled. Delivery prior to the commencement of induction with prostaglandin is easily explained by the natural history of pregnancy after intrauterine death. As mentioned in the introduction, spontaneous labour can be expected to occur in up to 90% of women within 14 days of fetal demise. Alternatively, if progesterone inhibition promotes uterine activity in the third trimester, as already demonstrated for the first half of pregnancy, trilostane alone may have contributed to the onset of labour in these women. In Study 1, as with the studies of Padayachee *et al*[46] and Cabrol *et al*[45], small numbers do not permit reliable conclusions.

Clearly, a group of women presenting with death of the fetus in utero is heterogeneous. The cause of death may originate in the fetal, placental or maternal compartments. It may be acute or preceded by a chronic period of fetal and/or maternal stress and the duration of intrauterine death may vary from less than 24 hours to more than five weeks. In our population, accurate determination of gestational age is often limited by the lack of a clear record of the last menstrual period, the recent use of injectable progestogen contraceptives, late booking and by the lack of early ultrasound dating. The duration of intrauterine death is equally difficult to determine with infrequent or non-attendance at antenatal clinics or when presentation is delayed due to a lack of information or access. The accuracy of clinical assessment at presentation is compromised further when the gestational age cannot be

calculated. In this study, the duration of intrauterine demise could not be estimated in 35% of women.

Analysis of the results failed to detect any correlation between treatment or induction-to-delivery intervals and gestational age, parity, apparent duration of intrauterine death (estimated in 65% of cases) or the pre-treatment Bishop's score. Support for these findings can be found in other third trimester studies that have used a similar induction regimen, with respect to parity, gestational age and duration of fetal death, and probably reflects the small numbers and heterogeneity of the trial populations.[32, 36, 52] The pre-treatment progesterone levels (ranked for gestational age) and the percentage fall in progesterone levels after 24 or 48 hours of trilostane were also unhelpful in the prediction of intervals to delivery.[83]

In this study, induction of labour with PGE_2 and oxytocin failed to procure delivery in 3 of 8 women in the placebo group and in 3 of 6 women in the trilostane group. This is in contrast to other studies of third trimester induction for intrauterine death where success rates (typically defined as delivery within 24 hours) have been reported of 92% for oxytocin alone[82] and from 94% to 100% with vaginal PGE_2 (± oxytocin).[32, 36, 37, 41, 52] Possible reasons for the contrast in success rate include the relatively low dose of prostaglandin used in this study, compared to those cited above, as well as a low-dose oxytocin regimen and a policy of avoiding amniotomy. A review by Ursell[82] described 113 cases of intrauterine death (beyond 22 weeks gestation and excluding cases of placental abruption) where labour was induced with either oxytocin alone or with a combination of oxytocin and amniotomy. He concluded that amniotomy decreased the interval to delivery and increased the probability of delivery within 24 hours. Although a slight increase in post-partum

pyrexia was observed in this group, the incidence was similar to that in a group of 50 patients in whom labour commenced spontaneously and was reportedly "easily managed with antibiotic therapy".

In previously cited studies,[32-38, 41, 42, 47-54] the mean induction-to-delivery interval has generally been shorter than was found in either group of this study. High prostaglandin doses have been characteristic of the earlier studies, particularly with vaginal administration. In contrast to the maximum 2 mg of PGE_2 vaginal gel used in Study 3, frequently repeated (2-4 hourly) bolus doses of 5, 10 and 20 mg PGE_2 of gel or pessary were typical. Due to small numbers in the reported trials, it is difficult to clearly assess the risk of major complications with larger prostaglandin doses. The anecdotal reports of uterine rupture, myocardial infarction and the risks of precipitate labour in these trials[36, 39-42] contrast with the relatively benign approach of expectant management and thus caution is advised with the use of prostaglandin in the third trimester. A reduction in the incidence of major complications by the use of low-dose prostaglandin regimens, however, will need to be assessed against the backdrop of prolonged or failed induction with its attendant increase in psychological and physical discomfort, infectious morbidity and the financial costs of inpatient and labour ward care.

Study power

Due to the wide range of induction-to-delivery intervals in these groups, much larger numbers would be required to detect a significant difference due to the use of trilostane. Based on the results of this study, it can be estimated that 66 women (33 in each group) would be required to detect a 50% reduction in induction-to-delivery interval (power 90%, significance 5%).[71] Much larger numbers would be required to detect smaller differences. Reducing the induction-to-delivery range, perhaps by a

more efficient induction regimen, would allow more accurate assessment of the efficacy of trilostane in this group.

Progesterone inhibition

Study 3 has clearly demonstrated that trilostane therapy causes a fall in progesterone levels in women with intrauterine fetal demise. The magnitude of this effect may be related to pre-treatment serum progesterone concentrations. In spite of a demonstrable effect and in contrast to gestations of less than 20 weeks duration, the lack of an obvious reduction in the induction-to-delivery interval with prostaglandin is disappointing.

As fetal demise may have a primary placental aetiology, placental function may have been deteriorating for a prolonged period prior to intrauterine death. Alternatively, a primary fetal cause of death may leave a healthy choriodecidual interface where placental activity continues for many days. Current theories of parturition favour a fetal role in its initiation and, as such, the death of the fetus may remove an important impetus towards the onset of labour.

Clearly, the factors influencing uterine contractility and sensitivity at different stages of pregnancy, and their relative importance is not fully appreciated. While there is ample evidence, from the effects of the progesterone-receptor antagonists, for the role of the progesterone receptor in the suppressive effect of progesterone on uterine contractility, the role of progesterone in the latter part of pregnancy and in the initiation of labour is unclear. Furthermore, the suspected role of the fetus in initiating parturition may begin many weeks before term, influencing the balance of suppressive and pro-contractile elements in the system – vis á vis Csapo's 'see saw' theory.[84] In the presence of fetal demise, the fetal contribution to this process is absent. If the

mechanism by which progesterone antagonists increase uterine sensitivity depends at some level on an intact fetoplacental unit then fetal demise would compromise the efficacy of such therapy. When fetal death occurs as a consequence of placental abruption, labour tends to follow shortly thereafter presumably as a consequence of the very high local concentrations of prostaglandin generated by the inflammatory process. When fetal demise occurs in the absence of inflammation, the mechanism by which prostaglandins are produced may be retarded by the absence of an intact fetoplacental unit. The effect of antiprogestin therapy in these circumstances would then differ from what is experienced with viable pregnancies. Further studies would be required to clarify whether increases in trilostane dose or duration or whether an increase in the dose of prostaglandin at induction would be more effective.

The short half-life of plasma progesterone[85, 86] explains the rapid fall in peripheral blood levels seen with progesterone synthesis inhibition. The magnitude of this effect may, however, be altered at the choriodecidual level due to uteroplacental haemodynamics and variations in metabolic rate. This may be a factor in the lack of correlation between the magnitude of decline in serum progesterone concentrations and induction-to-abortion intervals.

Placental activity after fetal death

It may be expected that progesterone levels would fall as a natural consequence of fetal death and it is interesting to note that, over the 48-hour period of treatment with placebo, this did not occur (Figure 14). It is known that the fetal contribution to progesterone synthesis is negligible and that the production of progesterone by the placenta is largely independent of uteroplacental perfusion, the precursor (maternal cholesterol) being readily available.[87]

Studies in animals and humans suggest that the clearance of progesterone from plasma following removal of the progesterone source (placental delivery, hysterotomy and luteectomy, oophorectomy) is not linear.[85, 88, 89] Following an initial rapid decline in progesterone levels, with a progesterone half-life of approximately 5 minutes in the first phase of decline, the elimination half-life increases considerably over the second and third phases.[85, 88, 89] In normal pregnancy, serum progesterone levels fall to 50% of delivery levels in the first hour after delivery of the placenta and then more gradually to less than 25% of delivery levels by 24 hours.[90] Lofgren and colleagues[91] calculated a mean elimination half-life for progesterone, following delivery, of approximately 38 minutes, which could possibly increase in obese women due to storage in and slow release from adipose tissue.[92] Expecting a relatively rapid clearance of progesterone from the plasma, it seems clear from the data in this study that placental production of progesterone has continued following fetal demise.

Blood levels of human placental lactogen (hPL) are related to placental function and with a short half-life (15 minutes) would fall rapidly if placental production ceased. Pre-treatment serum levels of hPL were within the normal ranges for gestational age in 13 of 19 women in Study 3, correlating with pre-treatment progesterone levels. This is further confirmation of continuing placental metabolic activity well beyond the time of fetal death.

Estriol – a role in parturition

Estriol is a placental product made almost exclusively from fetal adrenal dehydroepiandrosterone sulphate (DHEAS). Its synthesis therefore ceases when the fetus dies. Evidence of a role for estriol in the parturition process comes from its association with prolonged pregnancy in experiments of nature such as placental sulphatase deficiency and anencephaly.[93] The ratio of estriol to progesterone in viable

pregnancies has shown a positive correlation with spontaneous preterm labour (with intact membranes) and with the likelihood of spontaneous labour at term (versus the need for induction).[94, 95] In the presence of fetal demise, however, estriol levels are too low and too similar between women to offer any valuable means of distinction. This is not surprising when one considers its high rate of clearance from plasma.[96]

An interaction between estriol and placental corticotropin-releasing hormone in normal pregnancies has been suggested. This interaction may be regulated by the inhibitory effect of progesterone on the glucocorticoid receptor.[97] Increasing estriol in late pregnancy would promote activity of the placental 11β-hydroxysteroid dehydrogenase enzyme, which converts cortisol to inactive cortisone.[98] The result would be less fetal exposure to maternal cortisol and therefore less negative feedback on the fetal hypothalamic-pituitary-adrenal (HPA) axis. Increased fetal adrenal activity provides DHEAS as a substrate for increased estriol synthesis, and placental conversion of maternal cortisol to inactive cortisone is further augmented. Fetal cortisol production would also increase, competing with progesterone at placental glucocorticoid receptors and removing the suppressive effect on placental corticotropin-releasing hormone secretion. Placental corticotropin-releasing hormone may then stimulate the fetal pituitary further.[96]

In the case of fetal death, the postulated feedback mechanism between placenta and fetus promoting parturition (and possibly accelerated by the effects of fetal stress via the fetal HPA axis[99]) would be absent. The estriol:progesterone ratio would remain low favouring uterine quiescence and, without an intact fetal-placental circulation, some of the effects of an antiprogestin in promoting labour may be diminished.

An activated fetal HPA axis due to fetal stress prior to fetal demise may also play an important role in the likelihood of spontaneous labour or successful induction. With

the limitations of current scientific knowledge and of clinical information in most cases of non-placental causes of fetal death, quantification of such an effect would prove very difficult.

The mechanism of parturition and the relative roles of maternal, fetal and placental hormones in this process remain unclear. It seems obvious that the natural endocrine processes underlying the onset of labour may play a relatively minor role in some pathological states such as placental abruption, preterm rupture of membranes and chorioamnionitis. However, both a deeper understanding of the mechanism of human parturition, as well as knowledge of the status of all possible contributing factors in a given situation, will be required to allow individualisation and optimisation of care. Currently, obstetric management in which labour needs to be induced or suppressed remains unrefined and empirical.

CONCLUSION

Termination of pregnancy may be required at any gestation. Improving the efficiency of the service and effectiveness of the methods would reduce the significant maternal morbidity and mortality due to unsafe abortions. Medical methods of inducing abortion with prostaglandin are considerably more effective when combined with an antiprogestin. Mifepristone, the only antiprogestin preparation approved commercially for this indication, is currently unavailable in most countries due to licensing and cost considerations. Trilostane offers an effective, safe and less costly alternative to mifepristone.

This thesis has demonstrated the effectiveness, safety and acceptability of a low dose of trilostane in midtrimester termination of pregnancy. The high proportion of midtrimester pregnancy terminations in South Africa (and other developing countries) represents a significant maternal hazard and a strenuous demand on limited resources. A short and predictable interval to abortion and a reduction in pain and analgesia requirements are thus important benefits of an antiprogestin-prostaglandin regimen. The cost-effectiveness of a combination of trilostane and misoprostol would require that trilostane is administered in an outpatient setting prior to hospital admission for misoprostol induction of abortion. The shortened interval to abortion achieved with trilostane pre-treatment would reduce hospital stay by over 50% allowing many to be managed on a day-case basis. A further reduction in the induction-to-abortion interval may be achieved with higher misoprostol dose-regimens, increasing the proportion of women that can be effectively treated in a day-procedure unit. Studies are now required which can assess potential improvements to the regimen utilised in Study 1

and that will allow direct comparison with mifepristone-misoprostol and misoprostol-only regimens already in common use.

The findings in Study 1 support the idea of a threshold for progesterone suppression below which sensitisation of the uterus occurs. It is unclear whether this threshold is an absolute concentration of serum progesterone or rather a percentage of pre-treatment progesterone levels. Does the threshold vary with gestational age and between individuals and is it influenced by the rate of decline of progesterone concentrations? Further scientific study will be required to answer such questions.

A role for trilostane therapy in the second half of pregnancy has not been established in Studies 2 and 3. The success of pregnancy termination beyond twenty weeks gestation is highly unpredictable for reasons that remain obscure. Rather than the empirical approach that has an uncertain outcome, situations with diverse indications and circumstances require an individualised treatment plan. Larger clinical studies are required to compare different combinations of trilostane and misoprostol. An increase in the duration of trilostane therapy and/or misoprostol doses will be useful in clarifying the role of trilostane in later pregnancy and the optimal regimens among subgroups.

Induction of labour in the presence of intrauterine death may be difficult. The need to procure delivery without adding to the distress of bereavement requires an effective approach. Once again, individualisation of treatment is important as some women will labour readily with the minimum of stimulation and others will fail to respond to induction measures. Delaying induction for antiprogestin therapy may not confer benefit and may compound emotional distress in some situations. Currently there are no useful prognostic factors for predicting the success of induction.

In the presence of intrauterine death, both continued placental activity and the ability to suppress progesterone synthesis with trilostane have been clearly demonstrated in Study 3. This study has not been able, however, to identify a role for the use of trilostane in this clinical scenario. Heterogeneity is a significant limitation of a study of this kind and much larger numbers are required to allow identification and analysis of sub-groups. Limitations in the understanding of the normal physiology underlying parturition and of the pathophysiology relating to the various causes of intrauterine death preclude any useful explanation of the findings in this study. However, the importance of these findings may lie in the questions that they raise which can guide further scientific endeavour to a goal of improved obstetric care.

APPENDIX

Choice on Termination of Pregnancy Act, No 92 of 1996

The Termination of Pregnancy Bill was tabled in the South African parliament on 29 October 1996. It was promulgated as the Choice on Termination of Pregnancy Act, No 92 of 1996, effective from 3 February 1997.

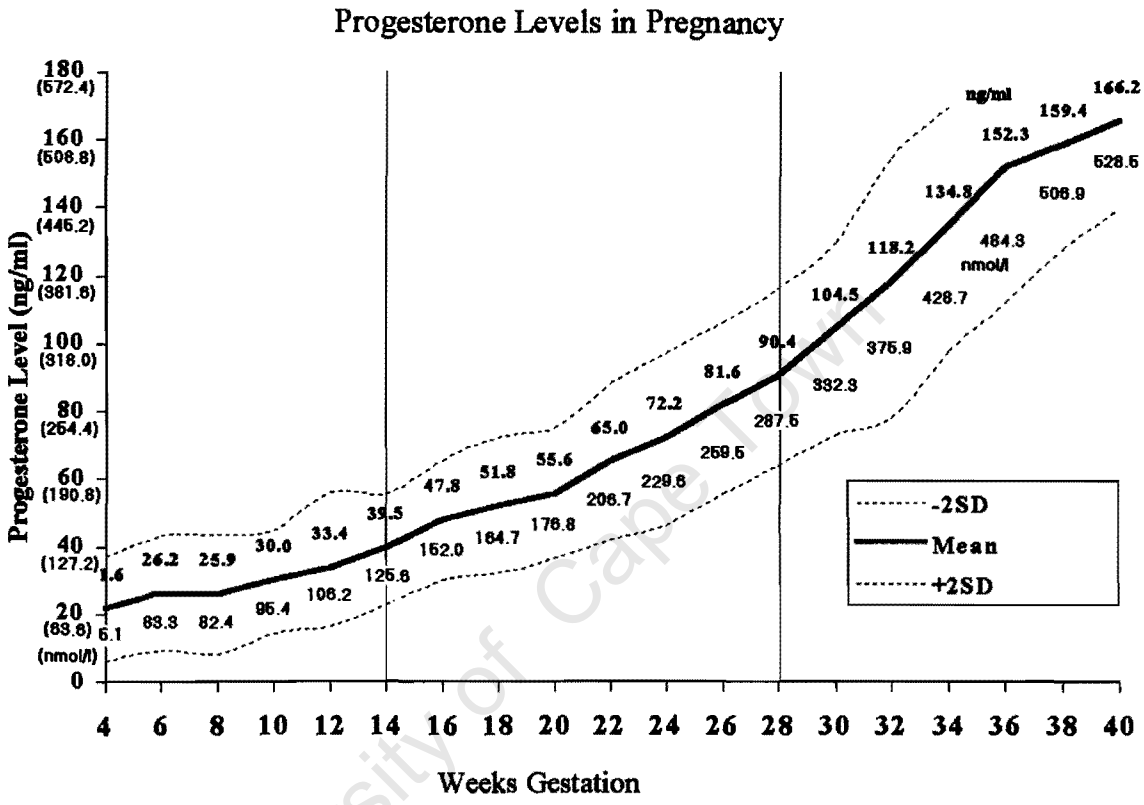
Under the Act, a woman has the right to choose to have an abortion during the first 12 weeks of pregnancy.

From the 13th to the 20th weeks of pregnancy, she may choose to have an abortion if a doctor is of the opinion that:

- The continued pregnancy is a risk to the woman's physical or mental health or,
- There is a substantial risk that the fetus would suffer a severe physical or mental abnormality, or
- The pregnancy resulted from rape or incest, or
- The pregnancy would severely affect the woman's social and economic circumstances.

After the 20th week of pregnancy, abortion can still take place if the pregnancy would endanger the woman's life or severely deform the fetus.

Serum progesterone in pregnancy - normal values.[100]



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STUDY NO:

TRILOSTANE MIDTRIMESTER DOSE-FINDING STUDY

CONSENT

I, ..., consent to participate in this study, which investigates methods of inducing abortion.

I understand that the study will involve the following:

1. Treatment with trilostane in addition to the usual administration of misoprostol to stimulate contractions
2. Medication will commence two (2) days prior to treatment with misoprostol
3. Blood samples will be taken twice daily for the duration of treatment
4. Additional therapy may be added should abortion not occur within 24 hours of starting the misoprostol

I understand that trilostane is unregistered medication which reduces hormonal production by the placenta and ovaries and is so doing sensitises the uterus to stimulation with medication that causes contractions.

I understand that this study is designed to evaluate possible improved therapy for termination of pregnancy.

I have received counselling with regard to my decision to undergo termination of pregnancy and I understand that participation in this study does not affect my choice in this matter.

I understand that I may withdraw from this study without prejudicing my management but I undertake first to discuss this with my medical attendants. I also understand that once treatment has commenced, there are possible long term consequences with regard to the development of the fetus, should I change my mind.

SIGNED:

PATIENT NAME:	**SIGNATURE:**	**DATE:**
.....	.....	.....
INVESTIGATOR:	**NAME:**	**SIGNATURE:**
	.....	.....
WITNESS:	**NAME:**	**SIGNATURE:**
	.....	.....

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STUDY NO:

TRILOSTANE TERMINATION OF PREGNANCY STUDY IN WOMEN WITH A FETAL ANOMALY

CONSENT

I, ..., consent to participation in this study, which investigates a new method of inducing labour.

I understand that the study will involve the following:

1. Treatment with ***either*** trilostane or inactive medication in addition to the usual administration of misoprostol to stimulate contractions
2. Medication will commence two (2) days prior to treatment with misoprostol
3. Blood samples will be taken twice daily for the duration of treatment
4. Additional therapy may be added should labour not occur within 24 hours of starting the misoprostol

I understand that trilostane is unregistered medication which reduces hormonal production by the placenta and ovaries and is so doing sensitises the uterus to stimulation with medication that causes contractions.

I understand that this study is designed to evaluate possible improved therapy for induction of labour

I have received counselling with regard to the problems in my baby, found on ultrasound examination, resulting in the advice to terminate the pregnancy

I understand I may withdraw from this study without prejudicing my management but I undertake first to discuss this with my medical attendants.

SIGNED:

PATIENT NAME:	**SIGNATURE:**	**DATE**
.....	.....	.....
INVESTIGATOR:	**NAME:**	**SIGNATURE:**
	.....	.....
WITNESS:	**NAME:**	**SIGNATURE:**
	.....	.....

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STUDY NO:

TRILOSTANE INDUCTION OF LABOUR STUDY IN WOMEN WITH INTRAUTERINE DEATH

CONSENT

I, ..., consent to participation in this study, which investigates a new method of inducing labour.

I understand that the study will involve the following:

1. Treatment with ***either*** trilostane or inactive medication in addition to the usual administration of prostaglandin gel followed by an intravenous infusion (drip) with Pitocin to stimulate contractions
2. Medication will commence two (2) days prior to treatment with prostaglandins
3. Blood samples will be taken twice daily for the duration of treatment
4. Additional therapy may be added should labour not occur within 24 hours of starting the gel

I understand that trilostane is unregistered medication which reduces hormonal production by the placenta and ovaries and is so doing sensitises the uterus to stimulation with medication that causes contractions.

I understand that this study is designed to evaluate possible improved therapy for induction of labour.

I have received counselling with regard to the complications in my pregnancy resulting in the death of my baby.

I understand I may withdraw from this study without prejudicing my management but I undertake first to discuss this with my medical attendants.

SIGNED:

PATIENT NAME:	**SIGNATURE:**	**DATE**
.....	.....	.....
INVESTIGATOR:	**NAME:**	**SIGNATURE:**
	.....	.....
WITNESS:	**NAME:**	**SIGNATURE:**
	.....	.....

Inhibition of Progesterone Secretion as an Interceptive Strategy.

Tregoning SK, Zinn PM, Van der Spuy ZM.

Abstract at the European Society of Human Reproduction and Embryology

15th Annual Meeting 1999.

Methodology

This study was conducted by the same research group responsible for the formulation of Study 1. It was completed in the two-year period preceding Study 1. The methodology of Study 1 was based on this work.

The study was a double blind randomised placebo-controlled trial. It consisted of a trilostane arm and a placebo arm. The study design, inclusion and exclusion criteria, recruitment methods, consent and randomisation was as described for Study 1 (pages 35, 39 and 40 respectively).

Initially trilostane was administered for 72h, amended to 48 hours after interval analysis (n=10). The initial trilostane dose was 2400 mg (over 72 hours in incremental divided doses - 480 mg, 960 mg, 960 mg), modified to 1440 mg with reduction of the treatment period to 48 hours (480 mg, 960 mg).

The misoprostol dose regimen was as described for Study 1 (page 36).

Results

48 women were recruited, 24 in each arm. Three were withdrawn - two due to side effects (trilostane) and one due to a protocol violation (placebo).

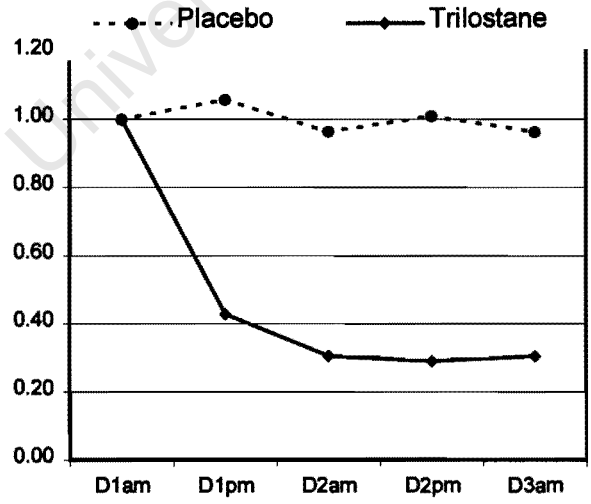
The mean induction to delivery interval was 9.2 hours (95% CI 7.3-11.1) in the trilostane arm and 21.6 hours (95% CI 15.4-27.8) in the placebo arm. This difference was significant, $P<0.01$.

Frequency of reported side effects.

Side effects	Trilostane (%)	Placebo (%)
Nausea &/or vomiting	9 (37)	4 (17)
Abdominal cramping	11 (46)	13 (54)
Itching eyes	6 (25)	0 (0)
Swollen tongue	3 (13)	0 (0)

Mean visual analogue scores (VAS) of acceptability as assessed by patients on a scale of 1 to 5. 1=minimum, 5=maximum, NS=not significant.

Acceptability	Trilostane VAS	Placebo VAS	Significance
Pain	2.23	4.41	P<0.01
Side effects	3.12	2.90	NS
Overall satisfaction	2.46	1.39	P<0.05



Mean serum progesterone levels, expressed as a percentage of pre-treatment levels, over 48 hours of treatment with either trilostane or placebo. D1am = Day 1 am.

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