# Day-specific probabilities of conception in spontaneous pregnancies

JULIEN J. STIRNEMANN[1,2], ADELINE SAMSON[2] JEAN-PIERRE BERNARD[1,3] AND JEAN-CHRISTOPHE THALABARD[2,4]

1. Department of Obstetrics and Maternal Fetal Medicine, Hôpital Necker-Enfants Malades, AP-HP, Université Paris Descartes, France
2. Applied Mathematics and Statistics, MAP5, UMR CNRS 8145, Université Paris Descartes, France
3. Centre Européen de Diagnostic et d'Exploration de la Femme (C.E.D.E.F.), Le Chesnay, France
4. Centre de Diagnostic, Hôtel- Dieu, AP-HP, Université Paris Descartes, France

RUNNING TITLE: Day-specific probabilities of conception

DISCLOSURES: none

IRB: none required

Abstract: 462 words

Text: 2946 words

Figures: 7

Tables: 3

Correspondance:

Julien J Stirnemann

Maternité et médecine materno-foetale, GHU Necker-Enfants Malades

149 rue de Sèvres, 75015 Paris

Tel : +33 1 44 49 40 30

Fax : +33 1 44 49 40 18

Email : j.stirnemann@gmail.com

ABSTRACT

**Title:** Day-specific probabilities of conception in spontaneous pregnancies

**Study question:** To estimate the measurement-error-free probability that conception occurs on a given day of the cycle, provided the cycle is fertile and the pregnancy is ongoing, using first trimester ultrasound fetal biometric measurements in spontaneous pregnancies

**Summary answer:** This study provides reference values of day-specific probabilities of conception in ongoing pregnancies. Moreover, these estimates could be used to refine the estimation of onset of pregnancy in the first trimester of pregnancy.

**What is already known:** The true date of conception is not observable and may only be estimated. Accuracy of these estimates impacts on obstetric management of ongoing pregnancies. Timing of ovulation and subsequent fertility has been extensively studied in prospective studies of non-pregnant fertile women using error-prone proxies such as hormonal changes, body-basal temperature and ultrasound, yielding day-specific probabilities of conception and fertile windows. In pregnant women, date of conception may be retrospectively estimated from early pregnancy ultrasound fetal measurement.

**Study design:** Retrospective analysis of a sample of pregnant women in the first trimester of pregnancy.

**Participants and methods:** The population comprised all consecutive pregnancies referred for a routine first trimester ultrasound, over a 3-year period (2009-2011) in a single ultrasound screening center (n=6323). Within this population, 5830 cases were selected for analysis. The date of conception was estimated using crown-rump length biometry. Day-specific probabilities were estimated across several covariates including age, cycle

characteristics and ethnicity, using deconvolution methods to account for measurement error.

**Main results:** Overall, the day-specific probability of conception sharply rises at 7 days, reaching its maximum at 15 days and returning to zero by 25 days. Older women tend to conceive earlier within their cycle as did women with regular cycles and White and Black women compared to Asian ethnicity. The probability of being within the fertile window closely matched previously published results from prospective monitoring studies of ovulation, with a 2% probability at day 4, a maximum probability of 58% at day 12 and a 5% probability by day 21 of the cycle.

**Limitations:** Although conception is believed to occur within hours following ovulation, a discrepancy is theoretically possible. However, when comparing our results to those of prospective studies, such a difference was not found. The equation used for estimating the date of pregnancy was estimated in IVF/ICSI pregnancies which could lead to potential bias in spontaneous pregnancies. However, in our population, the observed bias was negligible. Non-fertile cycles and early pregnancy losses are necessarily overlooked because of the nature of our data.

**Wider implications of the findings:** Because of the wider access to retrospective data and the potential bias in prospective studies of ovulation monitoring, this study should broaden the perspectives of future epidemiologic research in fertility and pregnancy monitoring.

**Study funding:** none

**Competing interests:** none

**Key-words:** timing of conception; fertile window; measurement error; ultrasound; early pregnancy;

## INTRODUCTION

Except in the specific case of assisted reproductive technologies such as in-vitro fertilization (IVF) or intracytoplasmic sperm injection (ICSI), the exact date of conception is unknown. Although ovulation generally occurs at around 14 days following the first day of last menses, a wide variation in the timing of ovulation has been found in prospective studies. Such studies generally rely upon hormonal changes (Wilcox *et al.*, 2000; Behre *et al.*, 2000; Cole *et al.*, 2009; Dunson *et al.*, 2001; Wilcox *et al.*, 1995; Venners *et al.*, 2006; O'Connor *et al.*, 2006; Dunson *et al.*, 1999), physiological changes such as basal body temperature (Royston, 1982; Royston *et al.*, 1984; Dunson *et al.*, 1999) or ultrasound (Marinho *et al.*, 1982; Ecochard *et al.*, 2001; Luciano *et al.*, 1990; Queenan *et al.*, 1980) to detect ovulation in healthy non-pregnant women monitored intensively in an experimental setting. However, although some may be more accurate than others, any indirect method aiming to detect ovulation or conception is prone to measurement-error (Dunson and Weinberg, 2000; Dunson *et al.*, 2001; Lynch *et al.*, 2006).

In pregnant women, the date of conception may be estimated from early fetal growth using sonographic biometry (Robinson, 1973). This method has been proved more reliable than last menstrual period for dating the onset of pregnancies (Mongelli and Gardosi, 1997; Gardosi and Geirsson, 1998; Gardosi *et al.*, 1997; Mustafa and David, 2001) and most national guidelines now consider early biometry as the method of choice for dating conception in routine practice (ACOG, 2009; NICE, 2008). However, dating of conception

using first trimester biometry remains an indirect observation of conception and therefore prone to error due to measurement error or biological variability in growth dynamics (Smith *et al.*, 2002, 1998).

Prospective estimation of date of ovulation in fertile women and retrospective estimation of date of conception in pregnant women are closely related since conception occurs within hours following ovulation, if ever (Royston, 1982; Wilcox *et al.*, 1995). Therefore, in pregnant women, the true day of conception may be safely considered as the true day of ovulation, although neither one is directly observed. Following, day-specific probabilities of conception are defined as the probability that conception occurs on a given day of the cycle (Lynch *et al.*, 2006), provided the cycle is fertile (see Appendix D for a formal presentation). Since a cycle may either be non-fertile or lead to an early loss, day-specific probabilities of conception should not be interpreted as the overall probability of clinical pregnancy.

Precise knowledge of the timing of conception, however, has important clinical implications: i) for counseling regarding fertility. In this context, hormonal ovulation monitoring methods have been made commercially available to help optimize the chances of conception (Behre *et al.*, 2000). ii) for the follow-up of pregnancies regarding growth monitoring, screening for birth defects and management of delivery. With regard to these clinical implications, the objective of this study is to provide estimates of day-specific probabilities of conception using ultrasound fetal biometry in the first trimester as a proxy in a large cohort of spontaneous singleton pregnancies. A specific statistical method is used to take into account the measurement error inherent to ultrasound estimates of date of conception (Stirnemann *et al.*, 2012; Comte *et al.*, 2011). The estimated distribution allows to calculate the day-

specific probability that conception occurs within a 'fertile window' as defined by (Wilcox *et al.*, 2000).

## METHODS

### Population and data collection.

*General inclusion criteria:* The overall population comprised all consecutive pregnancies referred for a routine first trimester ultrasound, over a 3-year period (2009-2011) in a single ultrasound screening center. In this center, women either self-refer or are referred by another practitioner without restriction regarding gestational age or indication. However, women are scheduled at around 12 weeks following last menstrual period (LMP) unless otherwise requested.

*General exclusion criteria:* All multiple pregnancies were excluded as well as patients referred on the basis of a specific condition (i.e. second-line examination, threatened miscarriage, acute pelvic pain or bleeding, fever or abnormal vaginal discharge). Pregnancies originating from assisted reproductive technologies were also excluded as their cycles may be disturbed by infertility treatment.

No further selection was made on the basis of ultrasound findings or measurements. Therefore, the study population is an unselected sample from the general population of spontaneous singleton pregnancies ongoing in the first trimester at 11-14 weeks. For the analysis of day-specific probabilities, patients with an unknown or uncertain date of LMP were excluded as well as those with amenorrhea or recent (<3 months) pregnancy, breastfeeding or prior contraception use without return to normal cycles.

In addition, an ancillary sample was composed of all the ongoing pregnancies originating from non-donor, non-frozen egg IVF or ICSI within the initial population. This ancillary sample was used for estimating a dating equation based upon crown-rump length, as explained later.

Demographic characteristics as well as information regarding cycle characteristics were collected upon referral and recorded prospectively. All the data including demographic characteristics, medical records and ultrasound results, were stored in a dedicated database (Astraia gmbh, Germany). Within this population, a proportion of women had an additional early first-trimester ultrasound on parental demand for psychological reassurance prior to the routine fetal assessment at 11-14 weeks. This subgroup of patients with two observations was handled specifically in the course of statistical modeling.

**Ultrasound measurements.** All ultrasound examinations were performed according to french national guidelines (CTE, 2005) and according to the guidelines of the Fetal Medicine Foundation (FMF, 2012). Dating of pregnancy was based upon crown-rump length (CRL). All ultrasound examinations were performed using a Voluson E8 (General Electric, GE Medical System Europe, Buc, France). Quality-control of ultrasound measurements is routinely performed in this pregnancy screening center and was ongoing throughout the study period, using standardized imaging quality assessments and scoring, statistical checks and external audits.

Ultrasound measurements together with the corresponding covariates were collected under the responsability of an obstetrician (JPB) as part of the routine follow-up and were stored in a clinical database with the patient's consent. The database was secondarily accessed only by JPB, who extracted anonymously the routine data, which were retrospectively analyzed in

the present study. All the data were manipulated according to the French regulation on both protection of privacy (law #2004-801,08/06/2004) and biomedical research (law# 2004-806, 08/08/2004).

**Unbiased ultrasound predictions of date of pregnancy.** All published reference dating equations displayed some bias when tested in the spontaneous conception as well as in the IVF/ICSI population. Although overall bias was found as small as 0.5 days with some equations – which is consistent with previous reports (Sladkevicius *et al.*, 2005) - it would have strongly hampered the final estimation of day-specific probabilities. Therefore, to rule out the impact of ultrasound prediction bias, in a preliminary analysis, we estimated a new dating equation based upon the IVF/ICSI dataset comprising 286 pregnancies with a median crown-rump length of 63.6 mm (interquartile range=55.1-68.8). The date of IVF/ICSI fertilization was considered as the date of conception. The final predictive model was the following equation with fetal age in days and CRL in mm, estimated using fractional polynomials (Royston & Sauerbrei 2008) (Appendix A):

$$\text{Age} = 21.564 + 2.224 \times \text{CRL} - 0.342 \times \text{CRL} \times \ln(\text{CRL}) \quad (1)$$

**Statistical analysis and correction for error-in-measurement.** The first day of the menstrual cycle was defined by the onset of menstrual bleeding. The date of pregnancy predicted from ultrasound measurements of crown-rump length was considered as a noisy observation of the true underlying date using an additive noise model given by equation (1), $Z=X+\varepsilon$ where Z is the observed time interval between LMP and the predicted date of pregnancy based upon CRL measurement, X is the unknown true time interval between LMP and true date of conception, i.e. without measurement error, and $\varepsilon$ is an unknown error. The probability

distribution function (p.d.f.) of X (true time since LMP) was estimated using nonparametric deconvolution methods that are described elsewhere (Stirnemann et al. 2012; Comte et al. 2011). The assumptions regarding the distribution of the error ε were checked (Appendix B). This estimation algorithm makes use of repeated measurements in the subset of pregnancies with an early first-trimester additional ultrasound to yield a smooth estimation of the error-free p.d.f of X. The estimated distribution provides day-specific probabilities defining the probability that a given day of the cycle is the true date of conception. Day-specific probabilities were calculated according to cycles reported as regular or irregular, according to maternal age groups, and ethnicity.

Finally, in the overall population, we used the previous estimation of day-specific probabilities to calculate the probability that a given day of the cycle falls within a 'fertile window', defined by the probability that a given day of the cycle falls within the 5-day window preceding conception(Wilcox *et al.*, 2000, 1995). All analyses were implemented in R v2.15.0 (R Development Core Team), using the ‘deamer’ library.

## RESULTS

Over the study period, 6323 women were referred for a routine ultrasound examination with a singleton spontaneous pregnancy. In this population, women reported their last menstrual period with certainty in 5830 (92%) cases. In 397 (6%) cases, women were uncertain or could not remember the date of their last menses. In 96 (2%) cases, the present pregnancy occurred shortly after a previous pregnancy, interruption of contraception or amenorrhea without return to normal cycles. Only women with a certain date of LMP were

selected for further analysis. The demographic characteristics of this population are presented in Table 1.

Routine ultrasound examinations were performed at a median 86 days following LMP (or equivalently 12 weeks and 2 days, interquartile range (IQR) = 85-89 days). Within the study population, 939 women had an additional early first-trimester ultrasound for psychological reassurance prior to the scheduled 11-14 weeks routine ultrasound. In this subgroup, the first ultrasound was performed at a median 57 days (IQR=51-64 days) following LMP. This subgroup was used to correct for measurement error in the estimation of day-specific probabilities (Appendix B).

Figure 1 presents the error-free estimates of day-specific probabilities of conception across the female cycle in the overall population. This distribution is right-skewed, showing a sharp rise from 7 days onwards, reaching its maximum of 13% at 15 days and decreasing to zero by 25 days following LMP.

**Maternal age.** Day-specific probabilities were calculated for the 3 groups of maternal age presented in Table 1. Figure 2 shows that the distribution is narrower and that pregnancies occur earlier in women aged >35. The maximum probability occurred at 15 days for women aged <25 and at 14 days for women aged >25. Furthermore women aged <25 displayed more variation with higher probabilities of onset of pregnancy around 21 days.

**Characteristics of female cycles.** Within the group with certain date of LMP, 5035/5830 (86%) reported regular cycles and 795/5830 (14%) reported irregular cycles. Compared to women with reportedly regular cycles, women with irregular cycles displayed more variation in timing of onset of pregnancy (Figure 3), with an increased likelihood of pregnancies occurring later in the cycle.

**Ethnicity.** Little difference in the day-specific probabilities of conception was found across ethnic groups as demonstrated by the overlap of distributions in Figure 4. However White women were found with the least variable dates of conception, whereas Asian women displayed the greatest variability mostly due to later onset of pregnancies in their third week.

Smoking status did not show any significant difference regarding the distribution of day-specific probabilities (data not shown). A numerical table of the day-specific probabilities plotted in Figures 1 to 4 is provided in Appendix C.

**Probability of falling within the fertile window.** The fertile window was defined by the five days preceding the day of conception. Figure 5 displays the probability that a given day of the cycle falls within this fertile window for each day of the cycle in the overall population. The probability of being within a fertile window rises from 2% on day 4 onwards and reaches 58% by day 12. By day 21, the probability falls down to 5%.

## COMMENT

### Using retrospective data from pregnant women for estimating day-specific probabilities of conception

This study provides reference values for the probability that conception occurs on a given day of the cycle, provided the cycle is fertile. A formal presentation of the relationship between day–specific probabilities in prospective and retrospective designs is presented in Appendix D. Although our results are related to prospective studies of timing of ovulation (O'Connor *et al.*, 2006; Dunson *et al.*, 1999; Venners *et al.*, 2006; Dunson *et al.*, 2001; Wilcox

*et al.*, 1995, 2000), they differ in several ways: i) We were interested in the date of conception rather than ovulation. A discrepancy in timing is likely although of little clinical relevance since fertilization is believed to occur within hours following ovulation (Royston, 1982; Wilcox *et al.*, 1995). Therefore, in our study, the day-specific probabilities of conception are a close approximation of the day-specific probabilities of ovulation estimated in a sample of fertile cycles leading to a clinical pregnancy; ii) Since we considered only pregnant women, our results are obviously conditional to the occurrence of a clinical pregnancy ongoing throughout the first trimester. Therefore, by design, only fertile cycles were selected, necessarily overlooking potentially non-fertile cycles. However, it has been hypothesized that the timing of ovulation does not impact on fertility nor on the probability that a given cycle will yield a pregnancy (Wilcox *et al.*, 2000). Conversely, the same authors suggest a relationship between late implantation and early pregnancy loss (Wilcox *et al.*, 1999). This effect is also overlooked by design in our study.

**Fetal biometry as a proxy for estimating the date of conception**

Using fetal biometry as a proxy for determining day-specific probabilities may raise concerns regarding potential bias and magnitude of measurement error compared to previously-used hormonal tests. Moreover, we used IVF/ICSI pregnancies to determine a dating equation, which could further limit the application of our dating equation, given the long-standing debate regarding growth disorders associated with IVF/ICSI (Le Bouc *et al.*, 2010; Dumoulin *et al.*, 2010; Eaton *et al.*, 2012). However, this dating method showed negligible bias (-0.02 day) in spontaneous pregnancies within the time-frame of first-trimester ultrasound. Furthermore, the magnitude of the error (sd = 1.52 day) was similar to reported precisions of

urinary hormonal detection of ovulation in optimal experimental settings (see Appendix B) (O'Connor et al. 2006; Dunson et al. 1999).

**Comparison with the results of prospective studies**

Most studies aiming to determine the timing of ovulation involve intensive longitudinal monitoring of women using study-specific diagnostic methods, which is likely to induce some selection bias. In contrast, our study uses routine cross-sectional clinical observations in a general population setting. Therefore our results are less likely to be prone to selection bias or to any impact of follow-up design on measurements, especially since observations are performed only after natural conception occurring outside a research setting. Furthermore, this allows for much larger samples and easier access to data than prospective experimental studies.

Regardless of these differences, our findings regarding the timing of the fertile window closely match those of previous reports. Indeed, our estimates (Figure 5) are strikingly similar to those reported by Wilcox *et al.* (2000): the maximum probability was reached by day 12, displaying a probability of 58% compared to the 54% probability reported by Wilcox et al. However, whereas our results showed a probability <1% by day 28 and onward, Wilcox *et al.* (2000) found a 4-6% probability remaining in the fifth week. Two independent hypotheses are likely to explain this difference: i) the estimates given by Wilcox *et al.* (2000) are not corrected for measurement error and a biased error (i.e. the mean error is not zero) could cause such an effect; ii) it may also be hypothesized that these late ovulations are non-fertile or at high risk of early pregnancy loss and therefore excluded in our data. Our results regarding the effect of maternal age and ethnicity are also consistent with previous reports showing a shortening of cycle length in women aged >35 and a longer cycle in Asian women

compared to White women (Liu *et al.*, 2004), although we acknowledge the difference between groups is small. Our results are also consistent with Wilcox *et al.* (2000), showing that conception occurs relatively later in women with reportedly irregular cycles compared to women with reportedly regular cycles.

**Clinical implications and relevance for epidemiology studies**

We wish to emphasize that achieving to describe the physiological variability in onset of pregnancy using simple routine clinical data in a large-scale sample should help broaden the perspectives of future research regarding the understanding of the relationship between physiological characteristics and fertility. Furthermore, with regard to the clinical implications of dating accuracy discussed in the Introduction, this study yields measurement-error free values for day-specific probabilities of conception according to several covariates which should be useful for fertility counseling as well as for dating pregnancy. In the context of fertility counseling, these estimates should be interpreted as the probability of conception provided the cycle is fertile and that the resulting pregnancy carries on through the first trimester. This definition may appear as a more pragmatic and clinically relevant concept than the probability of ovulation since it rules out the association between timing of ovulation and fertility and the association between timing of ovulation and early pregnancy loss. In the context of obstetric management of pregnancy, our results could help practitioners in refining the estimated date of pregnancy given by an early pregnancy ultrasound measurement. Reporting the date of conception predicted by a CRL equation within a table of day-specific probabilities provides some measure of the likelihood that this estimate, derived from an ultrasound measurement, is actually the true date of conception. Furthermore, reference values provided in Table C would allow prenatal care-providers to

implement maternal characteristics, such as age, cycle characteristics, ethnicity etc. in their appraisal of the most likely date of conception.

**Acknowledgments**

We thank Pr. Bouyer (Inserm, CESP, UMRS 1018 Université Paris Sud) for his insightful comments and his critical help in reviewing the final manuscript.

We thank Pr Ville (Obstetrics and Maternal – Fetal Medicine, Hôpital Necker-Enfants Malades, APHP and Université Paris Descartes) for his help in establishing the rationale for this study.

**Contribution to authorship**

Julien J Stirnemann initiated the study, analyzed the data, interpreted the results and wrote the manuscript.

Adeline Samson participated in the statistical analysis of the data and reviewed the manuscript.

Jean-Pierre Bernard leads the screening center and made the data available. He participated in the design as well as in the interpretation of the results.

Jean-Christophe Thalabard initiated the study, actively participated in the analysis and the interpretation of the results. He reviewed all versions of the manuscript.

# REFERENCES


ACOG. American Congress of Obstetricians and Gynecologists. Practice Bulletin No. 101: Ultrasonography in pregnancy. *Obstetrics and Gynecology* 2009; 113 (2 Pt 1): 451–461.

Behre HM, Kuhlage J, Gassner C, Sonntag B, Schem C, Schneider HP, Nieschlag E. Prediction of ovulation by urinary hormone measurements with the home use ClearPlan Fertility Monitor: comparison with transvaginal ultrasound scans and serum hormone measurements. *Human reproduction (Oxford, England)* 2000; 15 (12): 2478–2482.

Bland JM, Altman DG. Measuring agreement in method comparison studies. *Statistical Methods in Medical Research* 1999; 8 (2): 135–160.

Le Bouc Y, Rossignol S, Azzi S, Steunou V, Netchine I, Gicquel C. Epigenetics, genomic imprinting and assisted reproductive technology. *Annales d'endocrinologie* 2010; 71 (3): 237–238.

Cole LA, Ladner DG, Byrn FW. The normal variabilities of the menstrual cycle. *Fertility and Sterility* 2009; 91 (2): 522–527.

Comte F, Samson A, Stirnemann J. *Deconvolution estimation of onset of pregnancy with replicate observations*. Available from: http://hal.archives-ouvertes.fr/hal-00588235_v2/. 22 April 2011.

CTE. *Rapport du Comité national technique de l'échographie de dépistage prénatal*. Available from: http://www.ladocumentationfrancaise.fr/rapports-publics/054000356/index.shtml. 2005.

Dumoulin JC, Land JA, Van Montfoort AP, Nelissen EC, Coonen E, Derhaag JG, Schreurs IL, Dunselman GA, Kester AD, Geraedts JP, *et al.* Effect of in vitro culture of human embryos on birthweight of newborns. *Human reproduction (Oxford, England)* 2010; 25 (3): 605–612.

Dunson DB, Baird DD, Wilcox AJ, Weinberg CR. Day-specific probabilities of clinical pregnancy based on two studies with imperfect measures of ovulation. *Human Reproduction (Oxford, England)* 1999; 14 (7): 1835–1839.

Dunson DB, Weinberg CR. Modeling human fertility in the presence of measurement error. *Biometrics* 2000; 56 (1): 288–292.

Dunson DB, Weinberg CR, Baird DD, Kesner JS, Wilcox AJ. Assessing human fertility using several markers of ovulation. *Statistics in Medicine* 2001; 20 (6): 965–978.

Eaton JL, Lieberman ES, Stearns C, Chinchilla M, Racowsky C. Embryo culture media and neonatal birthweight following IVF. *Human reproduction (Oxford, England)* 2012; 27 (2): 375–379.

Ecochard R, Boehringer H, Rabilloud M, Marret H. Chronological aspects of ultrasonic, hormonal, and other indirect indices of ovulation. *BJOG: An International Journal of Obstetrics and Gynaecology* 2001; 108 (8): 822–829.

FMF. *The Fetal Medicine Foundation*. Available from: http://www.fetalmedicine.com/fmf/. 2012.

Gardosi J, Geirsson RT. Routine ultrasound is the method of choice for dating pregnancy. *British Journal of Obstetrics and Gynaecology* 1998; 105 (9): 933–936.

Gardosi J, Vanner T, Francis A. Gestational age and induction of labour for prolonged pregnancy. *British Journal of Obstetrics and Gynaecology* 1997; 104 (7): 792–797.

Liu Y, Gold EB, Lasley BL, Johnson WO. Factors affecting menstrual cycle characteristics. *American Journal of Epidemiology* 2004; 160 (2): 131–140.

Luciano AA, Peluso J, Koch EI, Maier D, Kuslis S, Davison E. Temporal relationship and reliability of the clinical, hormonal, and ultrasonographic indices of ovulation in infertile women. *Obstetrics and Gynecology* 1990; 75 (3 Pt 1): 412–416.

Lynch CD, Jackson LW, Buck Louis GM. Estimation of the day-specific probabilities of conception: current state of the knowledge and the relevance for epidemiological research. *Paediatric and Perinatal Epidemiology* 2006; 20 Suppl 13–12.

Marinho AO, Sallam HN, Goessens LK, Collins WP, Rodeck CH, Campbell S. Real time pelvic ultrasonography during the periovulatory period of patients attending an artificial insemination clinic. *Fertility and Sterility* 1982; 37 (5): 633–638.

Mongelli M, Gardosi J. Birth weight, prematurity and accuracy of gestational age. *International Journal of Gynaecology and Obstetrics: The Official Organ of the International Federation of Gynaecology and Obstetrics* 1997; 56 (3): 251–256.

Mustafa G, David RJ. Comparative accuracy of clinical estimate versus menstrual gestational age in computerized birth certificates. *Public Health Reports (Washington, D.C.: 1974)* 2001; 116 (1): 15–21.

NICE. *National Institute for Health and Clinical Excellence. Antenatal care*. Available from: http://www.nice.org.uk/. 26 March 2008. Guidance/Clinical Guidelines.

O'Connor KA, Brindle E, Miller RC, Shofer JB, Ferrell RJ, Klein NA, Soules MR, Holman DJ, Mansfield PK, Wood JW. Ovulation detection methods for urinary hormones: precision, daily and intermittent sampling and a combined hierarchical method. *Human Reproduction (Oxford, England)* 2006; 21 (6): 1442–1452.

Queenan JT, O'Brien GD, Bains LM, Simpson J, Collins WP, Campbell S. Ultrasound scanning of ovaries to detect ovulation in women. *Fertility and Sterility* 1980; 34 (2): 99–105.

R Development Core Team. *R: A Language and Environment for Statistical Computing*. Vienna, Austria, R Foundation for Statistical Computing.

Robinson HP. Sonar measurement of fetal crown-rump length as means of assessing maturity in first trimester of pregnancy. *British Medical Journal* 1973; 4 (5883): 28–31.

Royston JP. Basal body temperature, ovulation and the risk of conception, with special reference to the lifetimes of sperm and egg. *Biometrics* 1982; 38 (2): 397–406.

Royston JP, Humphrey SJ, Flynn AM, Marshall J, Zarzosa-Perez A. An automatic electronic device (Rite Time) to detect the onset of the infertile period by basal body temperature measurements. *British Journal of Obstetrics and Gynaecology* 1984; 91 (6): 565–573.

Royston P, Sauerbrei W. *Multivariable Model - Building: A Pragmatic Approach to Regression Anaylsis based on Fractional Polynomials for Modelling Continuous Variables*. 1st edition. Wiley.

Sladkevicius P, Saltvedt S, Almström H, Kublickas M, Grunewald C, Valentin L. Ultrasound dating at 12-14 weeks of gestation. A prospective cross-validation of established dating formulae in in-vitro fertilized pregnancies. *Ultrasound in Obstetrics & Gynecology: The Official Journal of the International Society of Ultrasound in Obstetrics and Gynecology* 2005; 26 (5): 504–511.

Smith GC, Smith MF, McNay MB, Fleming JE. First-trimester growth and the risk of low birth weight. *The New England Journal of Medicine* 1998; 339 (25): 1817–1822.

Smith GCS, Stenhouse EJ, Crossley JA, Aitken DA, Cameron AD, Connor JM. Early-pregnancy origins of low birth weight. *Nature* 2002; 417 (6892): 916.

Stirnemann JJ, Comte F, Samson A. Density estimation of a biomedical variable subject to measurement error using an auxiliary set of replicate observations. *Statistics in medicine* 2012;

Venners SA, Liu X, Perry MJ, Korrick SA, Li Z, Yang F, Yang J, Lasley BL, Xu X, Wang X. Urinary estrogen and progesterone metabolite concentrations in menstrual cycles of fertile women with non-conception, early pregnancy loss or clinical pregnancy. *Human Reproduction (Oxford, England)* 2006; 21 (9): 2272–2280.

Wilcox AJ, Baird DD, Weinberg CR. Time of implantation of the conceptus and loss of pregnancy. *The New England journal of medicine* 1999; 340 (23): 1796–1799.

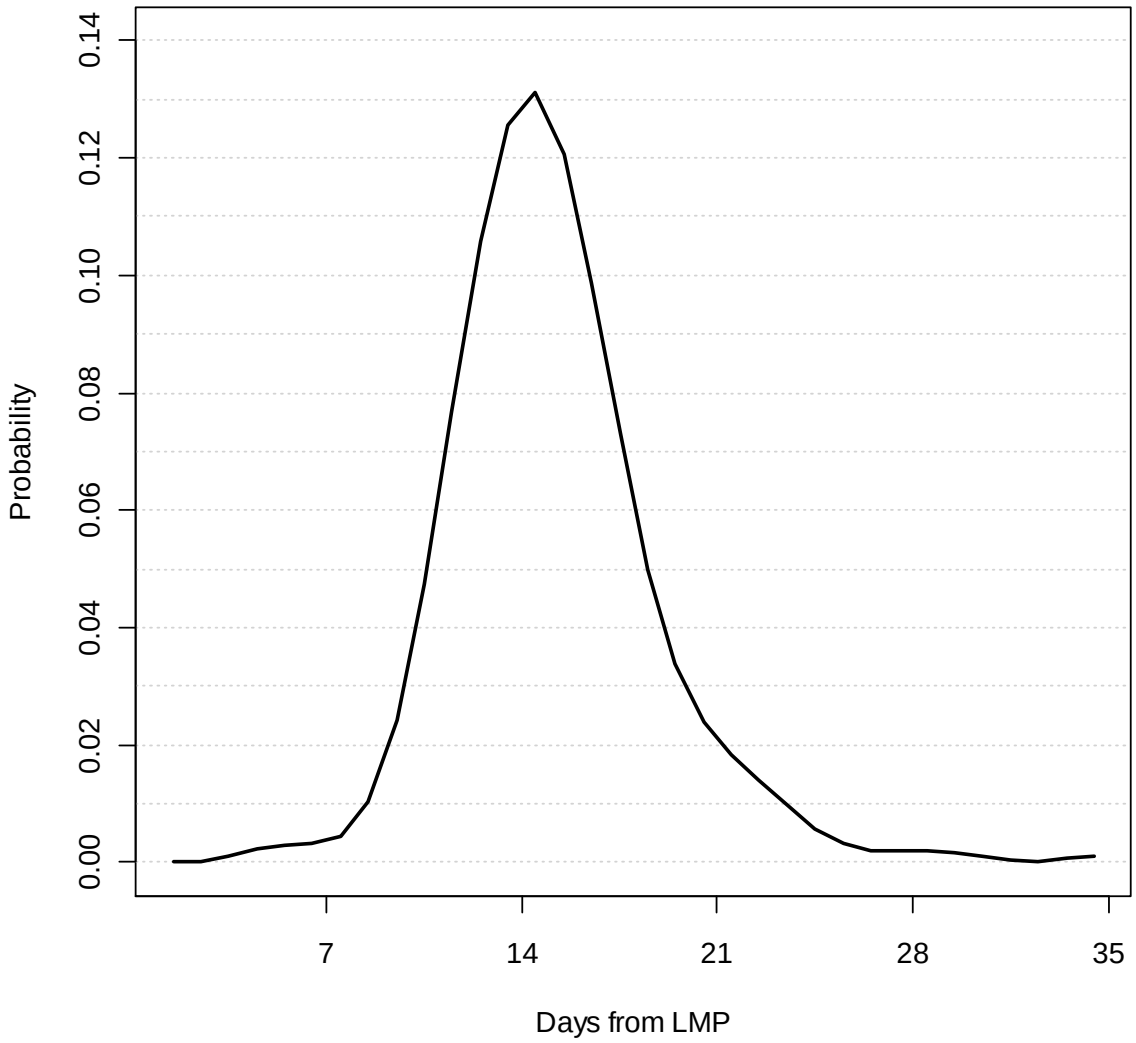
Wilcox AJ, Dunson D, Baird DD. The timing of the 'fertile window' in the menstrual cycle: day specific estimates from a prospective study. *BMJ (Clinical Research Ed.)* 2000; 321 (7271): 1259–1262.

Wilcox AJ, Weinberg CR, Baird DD. Timing of sexual intercourse in relation to ovulation. Effects on the probability of conception, survival of the pregnancy, and sex of the baby. *The New England Journal of Medicine* 1995; 333 (23): 1517–1521.

Table 1. Demographic characteristics of the study population (N=5830). Results are presented as N (%) unless otherwise specified. IQR: inter-quartile range; LMP: last menstrual period

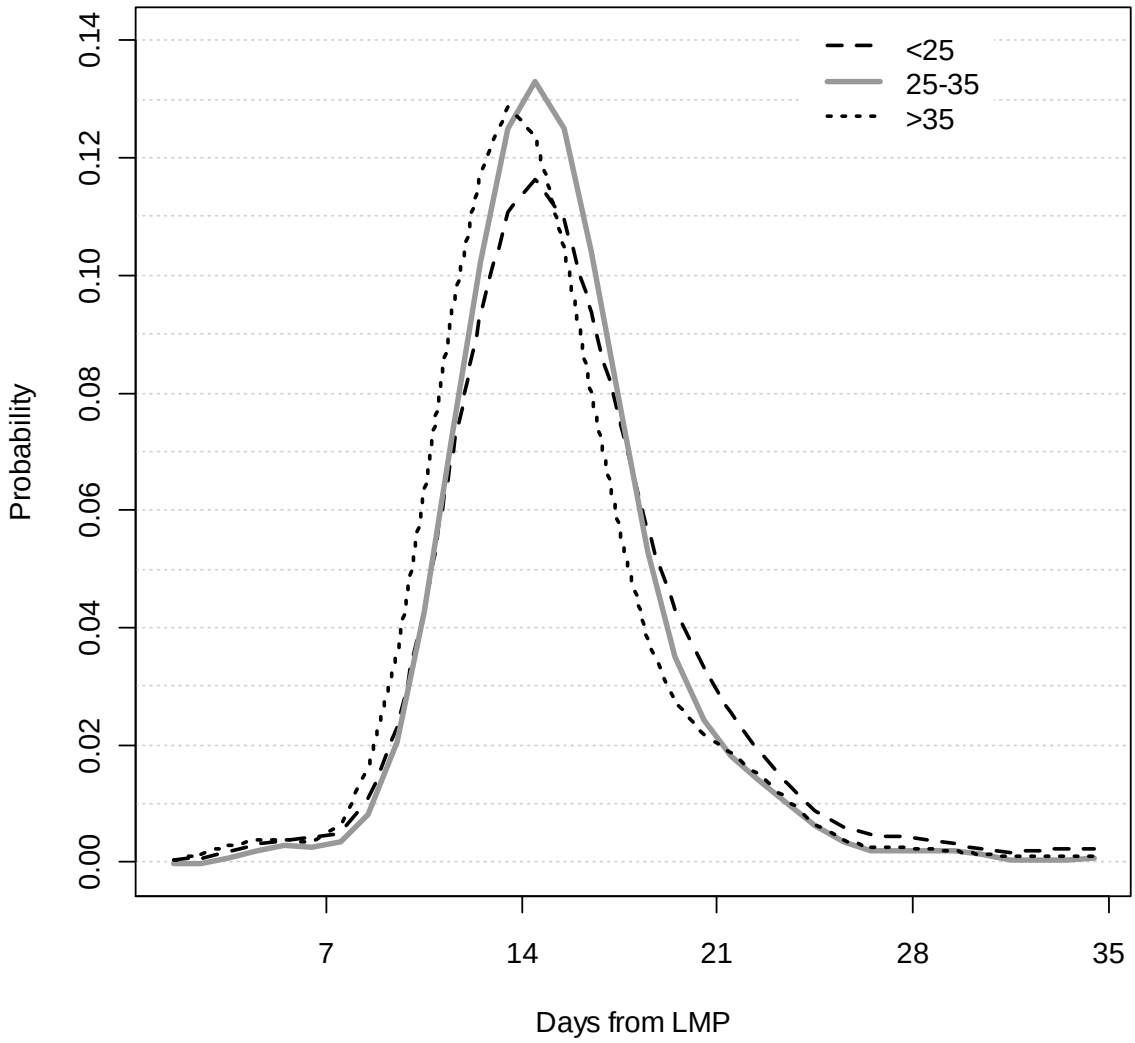
| Variable | N (%) |
|---|---|
| **Age** | |
| Median [IQR] | 30 [27-34] |
| < 25 | 614 (11) |
| 25-35 | 4042 (69) |
| >35 | 1173 (20) |
| **Nulliparous** | 3313 (57) |
| **Ethnicity** | |
| White | 5233 (90) |
| Black | 405 (7) |
| Asian | 154 (3) |
| Other | 38 (1) |
| **Smoking status** | |
| Non smoker | 4999 (86) |
| Stopped | 159 (3) |
| Smoker | 672 (12) |
| **Characteristics of last menstrual cycles** | |
| Regular | 5035 (80) |
| Irregular | 795 (13) |

1 Figure 1. Day-specific probabilities of conception in the overall population



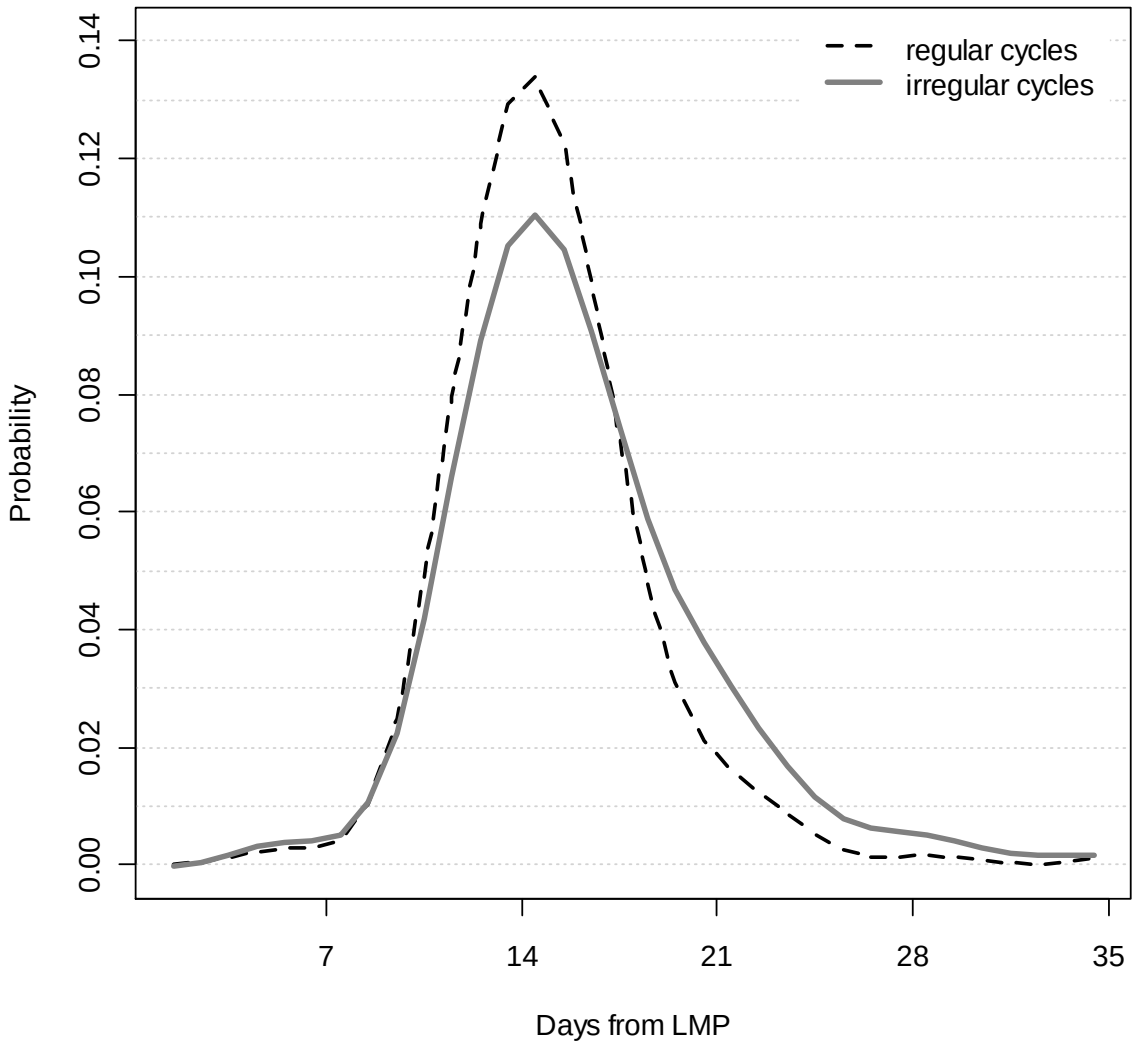

2

1 Figure 2. Day-specific probabilities of conception according to maternal age



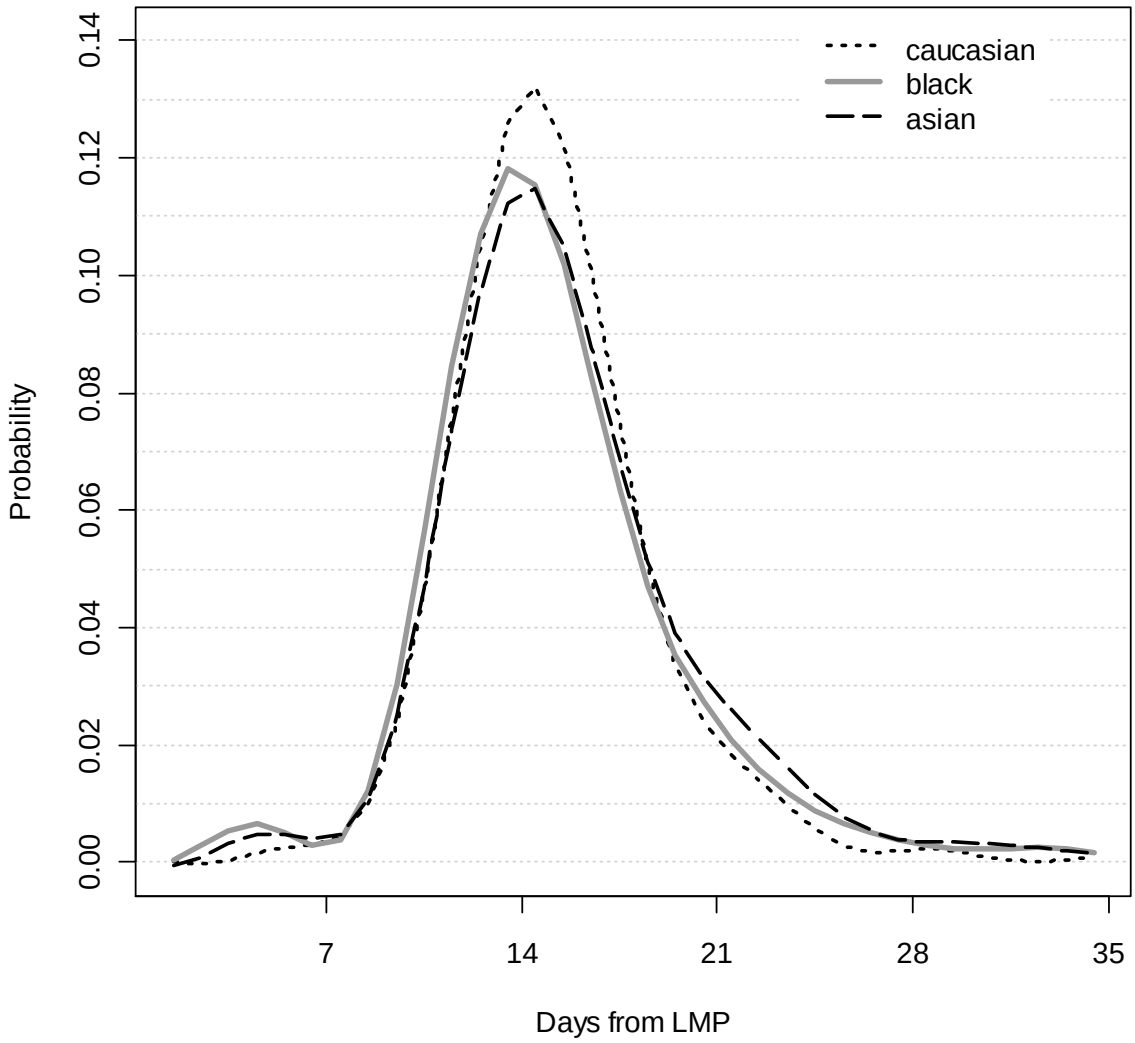

2

1 Figure 3. Day-specific probabilities of conception according to menstrual cycle characteristics



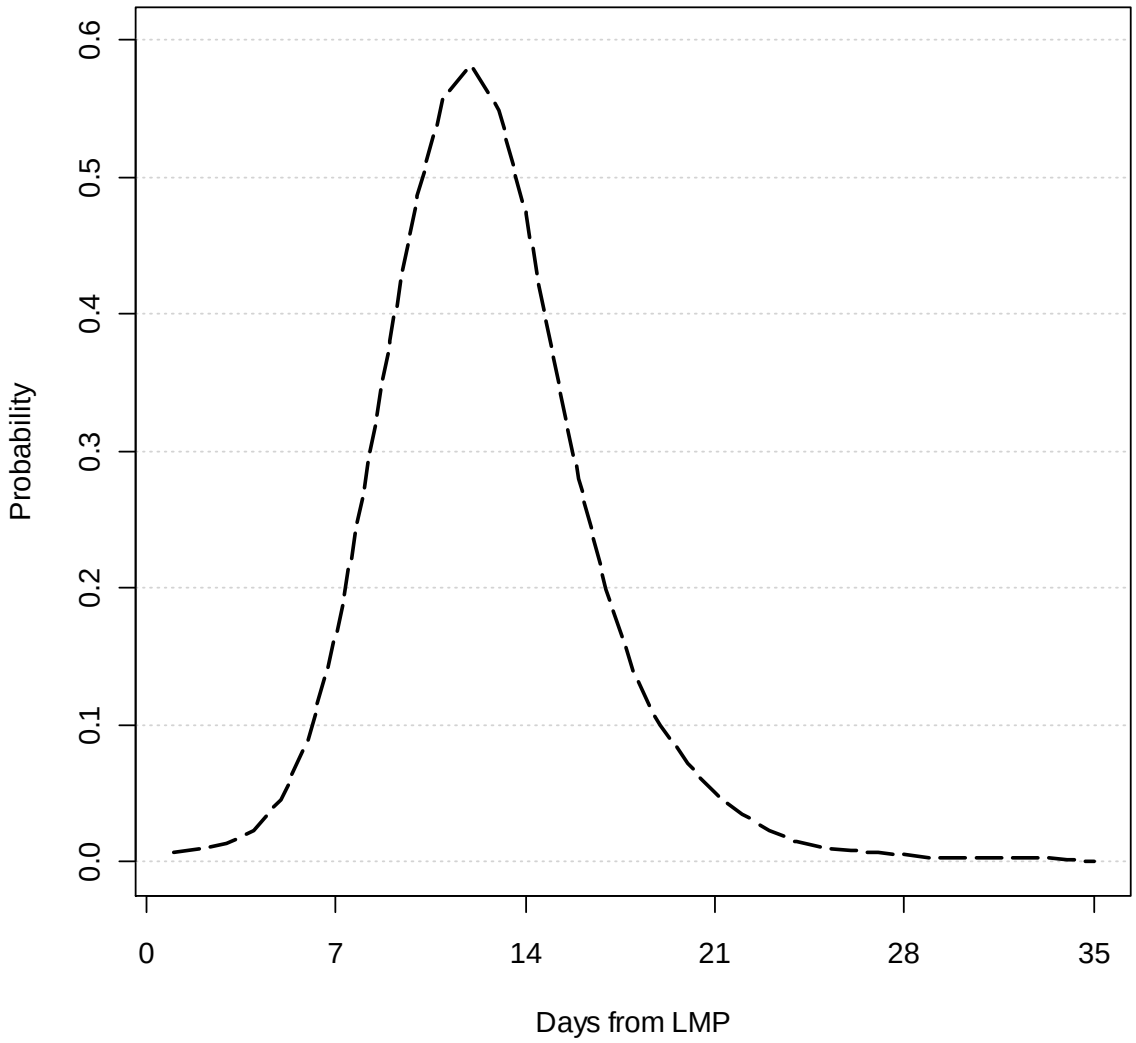

2

1 Figure 4. Day-specific probabilities of conception according to ethnicity




2

1 Figure 5. Day-specific probabilities of being within the 'fertile window'




2

3

## APPENDIX A. Dating equation in IVF/ICSI pregnancies

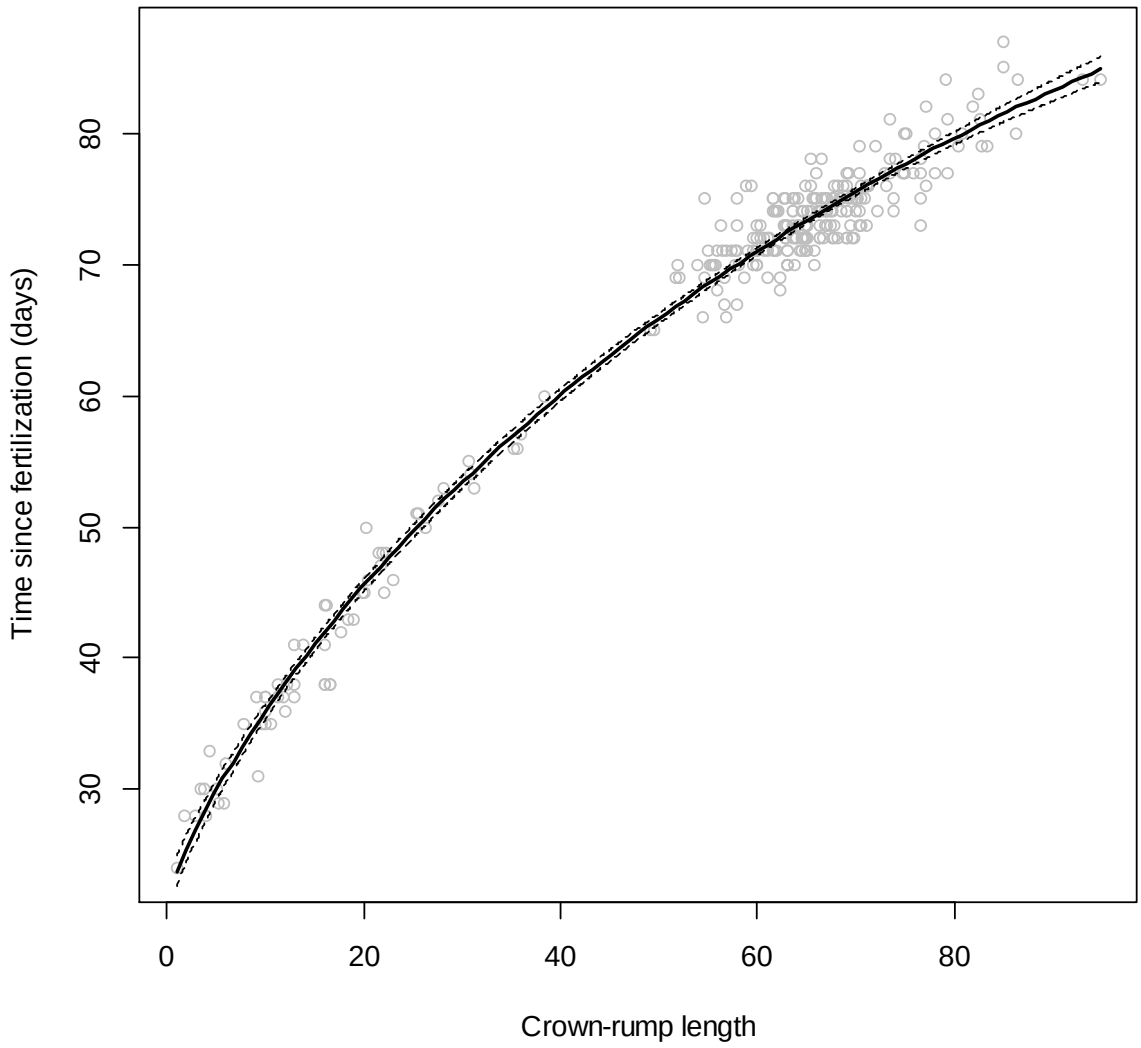
An equation for dating pregnancies according to first trimester crown rump length was derived in a population of 286 IVF/ICSI pregnancies. The age in days of the fetus since IVF/ICSI fertilization was modeled as a function of CRL. A linear fractional polynomial model was fitted using backward selection. The final model was the 2$^{nd}$ degree (df=4) fractional polynomial presented in Table A.

Table A. Estimates of the final model used for dating according to CRL in the IVF/ICS population

| Parameter | Estimate (standard error) | P-value |
|---|---|---|
| Intercept | 21.564 (0.67) | <0.001 |
| CRL | 2.224 (0.10) | <0.001 |
| CRL×ln(CRL) | -0.342 (0.02) | <0.001 |

The residual standard error was 1.98 days and $R^2$=0.982. Figure A presents the data together with the fitted model estimates. The Breusch-Pagan score test did not suggest heteroscedasticity in the residuals (P=0.43).

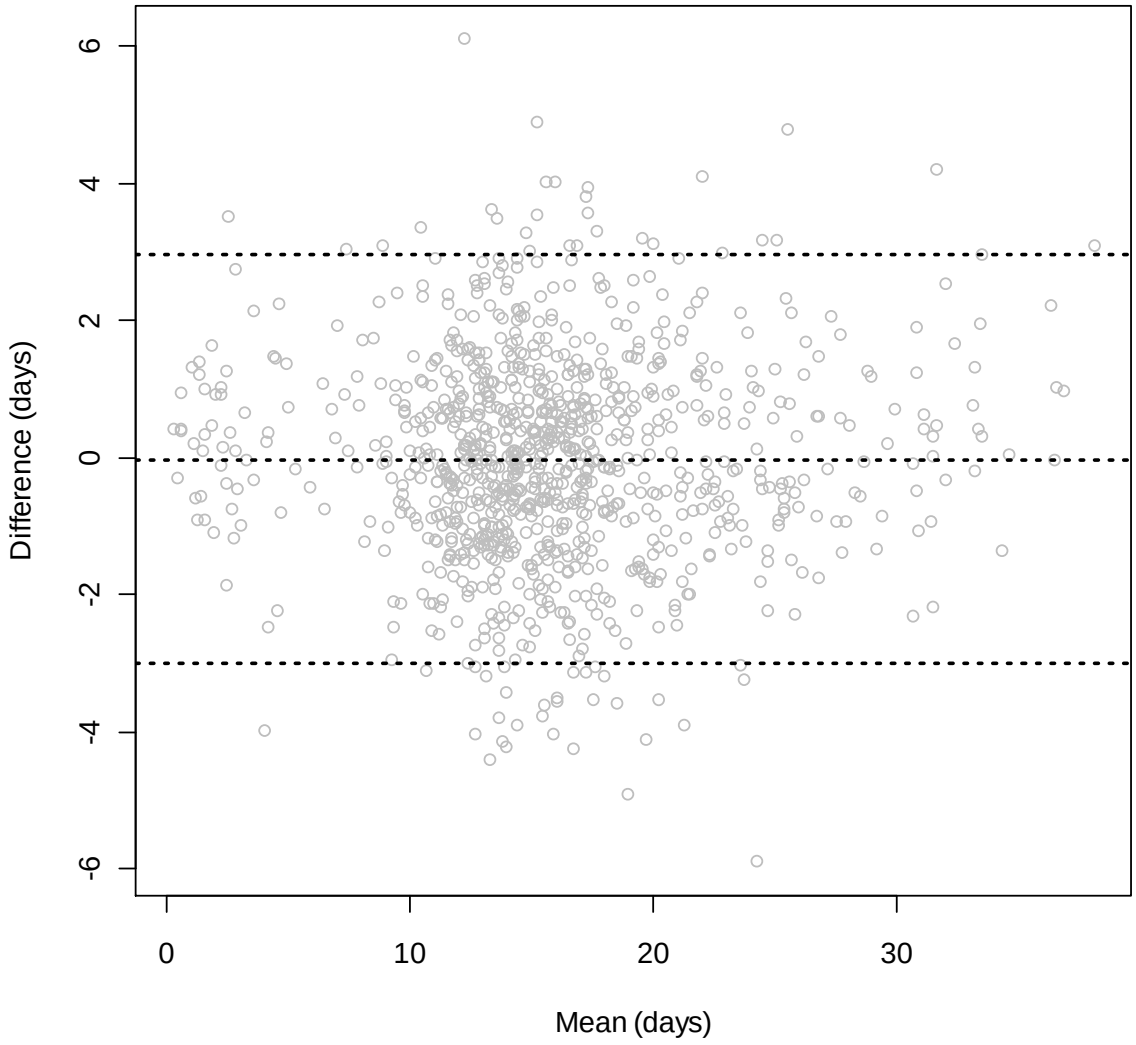
Figure A. Scatterplot of the time interval between ultrasound and fertilization versus crown-rump length in the IVF/ICSI population. The equation given by the model estimates is presented as a solid line and the 95% confidence band as dashed lines.



## APPENDIX B. Diagnostic plot of errors in longitudinal measurements of CRL

The deconvolution method that was used to estimate the probability density function of days of conception relies upon two underlying assumptions: i) the density of the error is symmetric around zero and ii) the errors are not heteroscedastic (the variance of the errors is constant). These assumptions were investigated in the group of spontaneous pregnancies with repeated ultrasound examinations. For each pair of observations Z1 and Z2 derived from repeated ultrasound, the difference was plotted against the mean of in the spirit of a Bland and Altman plot (Bland and Altman, 1999) (Figure B). The mean (SD) of the difference in predicted dates based upon CRL between longitudinal measurements was -0.02 (1.52) days. No trend was identified, nor was potential heteroscedasticity.

Figure B. Scatterplot showing the difference versus the mean of the two longitudinal observations in the subgroup with repeated measurements (Bland and Altman plot). The dotted lines indicate the bias and the 95% limits of agreement.



APPENDIX C.

Table C. Table of day-specific probabilities of conception in the population of women with an ongoing pregnancy after 11 weeks, according to maternal age groups, characteristics of cycles and ethnic groups (probability $\times$ 100).

| Day | Overall | Age | | | Cycles | | Ethnicity | | |
|---|---|---|---|---|---|---|---|---|---|
| | | <25 | 25-35 | >35 | Regular | Irregular | White | Black | Asian |
| 1 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |
| 2 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |
| 3 | 0.0 | 0.1 | 0.0 | 0.1 | 0.0 | 0.0 | 0.0 | 0.3 | 0.1 |
| 4 | 0.1 | 0.2 | 0.1 | 0.3 | 0.1 | 0.2 | 0.1 | 0.6 | 0.3 |
| 5 | 0.2 | 0.3 | 0.2 | 0.4 | 0.2 | 0.3 | 0.2 | 0.7 | 0.5 |
| 6 | 0.3 | 0.4 | 0.3 | 0.4 | 0.3 | 0.4 | 0.3 | 0.5 | 0.5 |
| 7 | 0.3 | 0.4 | 0.3 | 0.4 | 0.3 | 0.4 | 0.3 | 0.3 | 0.4 |
| 8 | 0.4 | 0.5 | 0.3 | 0.6 | 0.4 | 0.5 | 0.5 | 0.4 | 0.5 |
| 9 | 1.0 | 1.1 | 0.8 | 1.6 | 1.0 | 1.0 | 1.0 | 1.2 | 1.1 |
| 10 | 2.4 | 2.3 | 2.1 | 3.6 | 2.5 | 2.3 | 2.4 | 3.0 | 2.5 |
| 11 | 4.8 | 4.3 | 4.3 | 6.3 | 4.9 | 4.2 | 4.6 | 5.6 | 4.7 |
| 12 | 7.7 | 6.9 | 7.2 | 9.3 | 7.9 | 6.6 | 7.6 | 8.5 | 7.4 |
| 13 | 10.6 | 9.3 | 10.2 | 11.8 | 10.9 | 8.9 | 10.5 | 10.7 | 9.8 |
| 14 | 12.6 | 11.1 | 12.5 | 12.9 | 12.9 | 10.5 | 12.6 | 11.8 | 11.2 |
| 15 | 13.1 | 11.6 | 13.3 | 12.4 | 13.4 | 11.0 | 13.2 | 11.5 | 11.5 |
| 16 | 12.1 | 11.0 | 12.5 | 10.5 | 12.2 | 10.5 | 12.2 | 10.2 | 10.5 |
| 17 | 9.9 | 9.4 | 10.4 | 8.0 | 9.9 | 9.1 | 10.0 | 8.3 | 8.8 |
| 18 | 7.3 | 7.5 | 7.7 | 5.6 | 7.2 | 7.4 | 7.4 | 6.3 | 6.8 |
| 19 | 5.0 | 5.7 | 5.3 | 3.8 | 4.8 | 5.9 | 5.0 | 4.7 | 5.1 |
| 20 | 3.4 | 4.3 | 3.5 | 2.7 | 3.1 | 4.7 | 3.4 | 3.5 | 3.9 |
| 21 | 2.4 | 3.3 | 2.4 | 2.2 | 2.1 | 3.8 | 2.4 | 2.7 | 3.1 |
| 22 | 1.8 | 2.5 | 1.8 | 1.9 | 1.6 | 3.0 | 1.8 | 2.1 | 2.6 |
| 23 | 1.4 | 1.9 | 1.4 | 1.5 | 1.2 | 2.3 | 1.4 | 1.6 | 2.1 |
| 24 | 1.0 | 1.3 | 1.0 | 1.1 | 0.9 | 1.7 | 1.0 | 1.2 | 1.6 |
| 25 | 0.6 | 0.9 | 0.6 | 0.6 | 0.5 | 1.1 | 0.6 | 0.9 | 1.2 |
| 26 | 0.3 | 0.6 | 0.3 | 0.4 | 0.2 | 0.8 | 0.3 | 0.7 | 0.8 |
| 27 | 0.2 | 0.5 | 0.2 | 0.3 | 0.1 | 0.6 | 0.2 | 0.5 | 0.5 |
| 28 | 0.2 | 0.4 | 0.2 | 0.2 | 0.1 | 0.6 | 0.2 | 0.4 | 0.4 |
| 29 | 0.2 | 0.4 | 0.2 | 0.2 | 0.2 | 0.5 | 0.2 | 0.3 | 0.4 |
| 30 | 0.2 | 0.3 | 0.2 | 0.2 | 0.1 | 0.4 | 0.2 | 0.2 | 0.3 |
| 31 | 0.1 | 0.2 | 0.1 | 0.1 | 0.1 | 0.3 | 0.1 | 0.2 | 0.3 |
| 32 | 0.0 | 0.2 | 0.1 | 0.1 | 0.0 | 0.2 | 0.0 | 0.2 | 0.3 |
| 33 | 0.0 | 0.2 | 0.0 | 0.1 | 0.0 | 0.2 | 0.0 | 0.2 | 0.3 |
| 34 | 0.1 | 0.2 | 0.1 | 0.1 | 0.1 | 0.2 | 0.1 | 0.2 | 0.2 |
| 35 | 0.1 | 0.2 | 0.1 | 0.1 | 0.1 | 0.2 | 0.1 | 0.2 | 0.2 |

## APPENDIX D. Conditional probabilities

The ultimate goal of most prospective studies is to estimate the probability of a clinical pregnancy as a function of the day of cycle, timing of intercourse, number of cycles, time of implantation, demographic covariates and so forth. In this line, day-specific probabilities of clinical pregnancies may be defined as the conditional probability $p(clinical\ pregnancy|day\ of\ ovulation)$. In this manuscript we consider a different probability describing $p(day\ of\ ovulation|clinical\ pregnancy)$ which is the probability that a given day of the cycle is truly the day ovulation provided the cycle is ultimately fertile. This implies the hypothesis that conception occurs within a negligible time following ovulation.

Bayes' theorem yields the relationship between these two probabilities:

$$p(clinical\ pregnancy|day\ of\ ovulation) = \frac{p(day\ of\ ovulation|clinical\ pregnancy)p(clinical\ pregnancy)}{p(day\ of\ ovulation)} \quad (1)$$

Relationship (1) is not just formal: it shows that the "prospective" day-specific probability $p(clinical\ pregnancy|day\ of\ ovulation)$ and the "retrospective" day-specific probability $p(day\ of\ ovulation|clinical\ pregnancy)$ are directly related through clinically relevant information such as $p(clinical\ pregnancy)$, the probability of pregnancy per cycle and $p(day\ of\ ovulation)$, the probability that a given day of the cycle is the true day of ovulation regardless of subsequent outcome. Therefore, technically, one can compute "prospective" day-specific probabilities from "retrospective" day-specific probabilities provided values for $p(clinical\ pregnancy)$ and $p(day\ of\ ovulation)$.