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# First-trimester combined screening for trisomy 21 at 7–14 weeks' gestation

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**KEYWORDS:** combined screening; free β-hCG; first trimester; nuchal translucency; PAPP-A; screening for trisomy 21

## ABSTRACT

***Objective*** *To establish an algorithm for first-trimester combined screening for trisomy 21 with biochemical testing from 7 to 14 weeks' gestation and ultrasound testing at 11–13 weeks.*

***Methods*** *This was a multicenter study of 886 pregnancies with trisomy 21 and 222 475 unaffected pregnancies with measurements of free β-human chorionic gonadotropin (β-hCG) and pregnancy-associated plasma protein-A (PAPP-A) at 7–14 weeks' gestation. Multiple regression modeling of log-transformed marker values was used to produce log multiples of the median (MoM) values for PAPP-A and free β-hCG. The models included terms for the center attended and the machine used for biochemical analysis, gestational age, maternal racial origin, maternal weight, smoking status and method of conception. Bivariate Gaussian distributions were fitted to log MoM PAPP-A and log MoM free β-hCG in trisomy 21 and in unaffected pregnancies. In each case the patient-specific risk for trisomy 21 was estimated by multiplying the individual maternal age-related risk with the likelihood ratio (LR) for fetal nuchal translucency (NT) according to the mixture model and the combined LR for maternal serum free β-hCG and PAPP-A. Estimates of detection rates for trisomy 21 and false-positive rates were calculated for combined screening with measurements of NT at 12 weeks together with measurements of free β-hCG and PAPP-A from 8 to 13 weeks.*

***Results*** *In trisomy 21 pregnancies the mean log MoM free β-hCG increased linearly with gestation between 7 and 14 weeks, whereas the relation between log MoM PAPP-A and gestation was fitted by a quadratic equation such that the maximum separation between trisomy 21 and unaffected pregnancies occurs at 9–10 weeks. At a false-positive rate of 3% the detection rate of combined screening at 12 weeks was 86% and this increased to 90% by biochemical testing at 9 weeks and ultrasound scanning at 12 weeks. The detection rate increased to 92% by measuring PAPP-A at 9 weeks and free β-hCG at the time of the scan at 12 weeks.*

***Conclusion*** *The performance of first-trimester biochemical screening for trisomy 21 is best at 9–10 weeks rather than at 7–8 or 11–14 weeks. Copyright © 2010 ISUOG. Published by John Wiley & Sons, Ltd.*

## INTRODUCTION

Effective screening for trisomy 21 is provided by a combination of maternal age, fetal nuchal translucency (NT) thickness, maternal serum free β-human chorionic gonadotropin (β-hCG) and pregnancy-associated plasma protein-A (PAPP-A) at around 11–13 weeks' gestation[1]. In the assessment of patient-specific risks for trisomy 21 the *a-priori* maternal age-related risk is multiplied by likelihood ratios (LR), determined from the deviation of the measured NT, free β-hCG and PAPP-A from the respective expected levels.

A study of trisomy 21 cases and controls across the first and second trimesters reported that there is a temporal variation of biochemical markers in cases with trisomy 21[2,3]. The temporal variation has two consequences for

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risk assessment: firstly, the classical constant median separation models lead to inappropriate risk estimates and there is a requirement for the use of variable median separation models; and secondly, the improved clinical discrimination of PAPP-A in earlier weeks may result in better performance of biochemical screening from blood sampling before 11 weeks. It was on the basis of this early analysis of temporality that the previous Fetal Medicine Foundation algorithms were based. Extensive data on serum biochemistry at 11–13 weeks' gestation suggest that the performance of screening is better when conducted at 11 than at 13 weeks. This occurs because, although the separation between trisomy 21 and unaffected pregnancies in serum free β-hCG increases with gestation, the decreasing separation in PAPP-A levels dominates[4].

In this study we present the combined data from four European centers containing 886 pregnancies with trisomy 21 and about 220 000 unaffected pregnancies with measurements of free β-hCG and PAPP-A at between 7 and 14 weeks' gestation.

## PATIENTS AND METHODS

This study is based on data on singleton pregnancies from prospective first-trimester combined screening for trisomy 21 at Center A (King's College Hospital, London; Fetal Medicine Centre, London), Center B (King George Hospital, Goodmayes; Harold Wood Hospital, Romford; Queens Hospital, Romford; Canterbury Hospital, Canterbury; William Harvey Hospital, Ashford; Queen Elizabeth The Queen Mother's Hospital, Margate; Homerton University Hospital, Hackney; St George's Hospital, Tooting; Epsom Hospital, Epsom; St Helier Hospital, Carshalton), Center C (Aarhus University Hospital, Skejby, Denmark; Regionshospitalet, Silkeborg; Regionshospitalet Randers and Regionshospitalet, Viborg, Denmark) and Center D (Fetal Medicine Centre, Nicosia, Cyprus and Alpha Medical Diagnosis, Limassol, Cyprus). Karyotype results and details on pregnancy outcomes were added into the databases as soon as they became available. The data from the UK on 96 803 unaffected pregnancies and 491 cases of trisomy 21 and from Denmark on 10 243 unaffected and 97 trisomic pregnancies have been included in previous publications[4,5].

Prospective assessment of the risk for trisomy 21 was based on a combination of maternal age, fetal NT thickness, and maternal serum PAPP-A and free β-hCG. Serum samples collected at between 7 and 14 weeks' gestation were analyzed to determine concentrations of free β-hCG and PAPP-A using automated machines that provide reproducible results within 30 min (Kryptor system, Brahms AG, Berlin, Germany, or Delfia Express system, Perkin Elmer, Waltham, MA, USA). Transabdominal and, when necessary due to impaired visualization, transvaginal, ultrasound NT measurements were performed at around 11–13 weeks' gestation to diagnose any major fetal defects and for measurement of the fetal crown–rump length (CRL) and NT thickness[1]. Gestational age was based on the CRL at the time of the NT measurement[6].

Maternal weight, measured at the time of serum screening, demographic characteristics, ultrasonographic measurements and biochemical results were recorded in computer databases. Racial origin was assessed by self-classification and included Caucasian, African, South Asian (Indian, Pakistani, Bangladeshi), East Asian (Chinese, Korean, Japanese) and mixed, which mainly consisted of those of Caucasian and Afro-Caribbean/African origin. Smoking status was based on a self-completed questionnaire, and each woman was classified as either a non-smoker or a smoker. The mode of conception was either spontaneous or with ovulation-induction drugs or assisted reproduction by *in-vitro* fertilization. Missing data on smoking, ethnicity or conception were accommodated by including a 'missing' factor for these variables. In a small proportion of cases (1.5%) the maternal weight was missing and these cases were allocated the mean of the women examined at the same center.

Multiple regression modeling[7] of log-transformed marker values was used to produce log multiples of the median (MoM) values for PAPP-A and free β-hCG. The models included terms for the center attended and machine used for biochemical analysis, gestational age, maternal racial origin, maternal weight, smoking status and method of conception. The mean log MoM values for free β-hCG and PAPP-A in trisomy 21 were represented as a linear and a quadratic function of gestational age. Bivariate Gaussian distributions were fitted to log MoM PAPP-A and log MoM free β-hCG in trisomy 21 and in unaffected pregnancies. The adequacy of the fitted model was assessed by inspecting histograms of the squared Mahalanobis distances with the chi-squared distribution superimposed. Gross outliers beyond the 99.99$^{th}$ centile were removed.

In each case the patient-specific risk for trisomy 21 was estimated by multiplying the individual maternal age-related risk by the LR for fetal NT according to the mixture model[8] and the combined LR for maternal serum free β-hCG and PAPP-A. Crude, standardized and model-based detection rates and false-positive rates were obtained by taking the proportion of cases with risks above a given risk threshold. The crude performance of screening refers to the observed values in our dataset. Standardized performance of screening was estimated after adjustments to take into account the maternal age distribution of pregnancies in England and Wales in 2000–2002[9]. Model-based estimates of screening performance were derived by examining simulated data from 500 000 unaffected pregnancies and 500 000 trisomy 21 pregnancies with the maternal age distribution of pregnancies in England and Wales in 2000–2002, fetal NT distributions according to the mixture model and log MoM free β-hCG and log MoM PAPP-A distributions as modeled from data in this study. Bootstrapping techniques with 100 replications were used to estimate

95% confidence intervals for standardized detection rates. Model-based estimates of detection rates for trisomy 21 and false-positive rates were also calculated for combined screening by maternal age, fetal NT according to the mixture model and serum free β-hCG and PAPP-A for measurements of NT at 12 weeks together with measurements of both free β-hCG and PAPP-A at 8, 9, 10, 11, 12 and 13 weeks as well as measurement of free β-hCG and PAPP-A separately at 8, 9, 10, 11, 12 and 13 weeks.

## RESULTS

The dataset included 222 475 euploid pregnancies and 886 trisomy 21 pregnancies. Details of maternal and fetal characteristics are shown in Table 1. On the basis of the maternal age distribution of the study population the estimated number of trisomy 21 pregnancies, at the time of screening, was 936 (95% prediction interval, 876–996).

**Table 1** Characteristics of the study population

| *Parameter* | *Value* |
|---|---|
| Center | |
| A | 56 771 (25.4) |
| B | 127 611 (57.1) |
| C | 11 665 (5.2) |
| D | 27 314 (12.2) |
| Maternal characteristics | |
| Maternal age (years) | 31.9 (27.7–35.8) |
| Maternal weight (kg) | 65.0 (58.0–73.8) |
| Mode of conception | |
| Spontaneous | 217 352 (97.3) |
| *In-vitro* fertilization | 5089 (2.3) |
| Ovulation-induction drugs | 647 (0.3) |
| Not reported | 273 (0.1) |
| Smoking status | |
| Smoker | 23 864 (10.7) |
| Non-smoker | 182 043 (81.5) |
| Not reported | 17 454 (7.8) |
| Ethnicity | |
| Caucasian | 189 819 (85.0) |
| African | 11 043 (4.9) |
| South Asian | 15 391 (6.9) |
| East Asian | 1755 (0.8) |
| Mixed | 3777 (1.7) |
| Not reported | 1576 (0.7) |
| Gestational age when tested | |
| 7 weeks | 283 (0.1) |
| 8 weeks | 6884 (3.1) |
| 9 weeks | 16 179 (7.2) |
| 10 weeks | 14 215 (6.4) |
| 11 weeks | 33 512(15.0) |
| 12 weeks | 98 011(43.9) |
| 13 weeks | 52 163 (23.4) |
| 14 weeks | 2114 (0.9) |
| Karyotype | |
| Normal | 222 475 (99.6) |
| Trisomy 21 | 886 (0.4) |

Data are given as *n* (%) or median ($25^{th}$–$75^{th}$ quartile).

### Distribution of log MoM free β-hCG and PAPP-A in unaffected and trisomy 21 pregnancies

In the unaffected pregnancies, the median gestational age-related concentrations of free β-hCG and PAPP-A are shown in Figure 1. The fitted regression relationships on gestational age (GA, days) and maternal weight (Wt, kg) in unaffected pregnancies were:

$$\begin{aligned}\text{Log}_{10}\ \text{free β-hCG} = &-3.23998\\ &-(0.0509672 \times (\text{GA} - 77))\\ &+(0.000448023 \times (\text{GA} - 77)^2)\\ &+(3.151635 \times \log_{10}(\text{GA} - 40))\\ &-(0.004502895 \times (\text{Wt} - 69))\\ &+(0.00002674235 \times (\text{Wt} - 69)^2)\end{aligned}$$

and

$$\begin{aligned}\text{Log}_{10}\ \text{PAPP-A} = &\ 0.194961\\ &+(0.0284402 \times (\text{GA} - 77))\\ &-(0.000352202 \times (\text{GA} - 77)^2)\\ &+(0.0000124371 \times (\text{GA} - 77)^3)\\ &-(0.00730782 \times (\text{Wt} - 69))\\ &+(0.0000292028 \times (\text{Wt} - 69)^2).\end{aligned}$$

Multiple regression analysis demonstrated significant effects on the measured concentrations from the center attended and machine used for biochemical analysis, gestational age and maternal racial origin, weight, smoking status and method of conception (Table 2). The effects are similar to our previously published estimates[1]. The MoM values for free β-hCG and PAPP-A

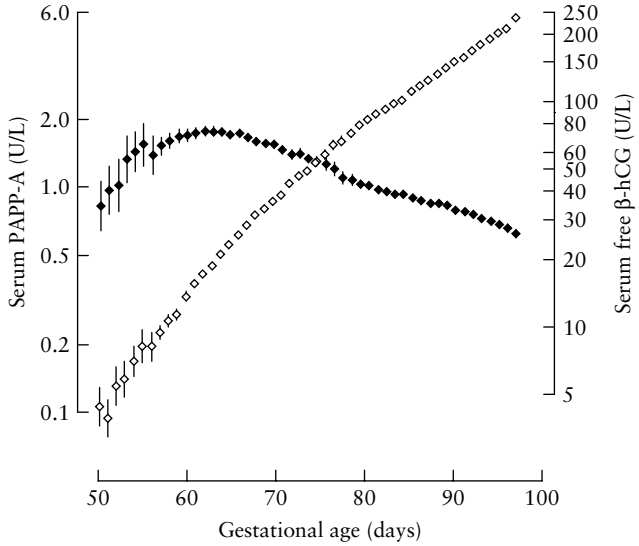


**Figure 1** Daily median concentrations with 95% CIs of pregnancy-associated plasma protein-A (PAPP-A) (◇) and free β-human chorionic gonadotropin (β-hCG) (◆).

**Table 2** Parameter estimates for the fitted Gaussian distributions for pregnancy-associated plasma protein-A (PAPP-A) and free β-human chorionic gonadotropin (β-hCG) in euploid pregnancies in the study

| | *PAPP-A* | | *Free β-hCG* | |
|---|---|---|---|---|
| | *Log scale* | *Original scale* | *Log scale* | *Original scale* |
| *Parameter* | *Estimate (95% CI)* | *Estimate (95% CI)* | *Estimate (95% CI)* | *Estimate (95% CI)* |
| Ethnicity | | | | |
| Caucasian | 0.0000 | 1.0000 | 0.0000 | 1.000 |
| African | 0.1919 (0.1874 to 0.1963) | 1.5555 (1.5396 to 1.5716) | 0.0488 (0.0438 to 0.0538) | 1.119 (1.106 to 1.132) |
| South Asian | 0.0170 (0.0132 to 0.0209) | 1.0400 (1.0308 to 1.0492) | −0.0522 (−0.0565 to −0.0479) | 0.887 (0.878 to 0.896) |
| East Asian | 0.0371 (0.0265 to 0.0478) | 1.0893 (1.0628 to 1.1164) | 0.0189 (0.0070 to 0.0309) | 1.045 (1.016 to 1.074) |
| Mixed | 0.0249 (0.0176 to 0.0322) | 1.0590 (1.0413 to 1.0771) | −0.0081 (−0.0163 to 0.0001) | 0.982 (0.963 to 1.000) |
| Smoking status | | | | |
| Non-smoker | 0.0000 | 1.0000 | 0.0000 | 1.000 |
| Smoker | −0.0755 (−0.0786 to −0.0724) | 0.8404 (0.8344 to 0.8465) | −0.0159 (−0.0194 to −0.0124) | 0.964 (0.956 to 0.972) |
| Mode of conception | | | | |
| Spontaneous | 0.0000 | 1.0000 | 0.0000 | 1.000 |
| IVF | −0.0383 (−0.0447 to −0.0320) | 0.9155 (0.9022 to 0.9290) | 0.0283 (0.0212 to 0.0354) | 1.067 (1.050 to 1.085) |
| Ovulation drugs | −0.0415 (−0.0591 to −0.0239) | 0.9089 (0.8728 to 0.9465) | 0.0218 (0.0020 to 0.0415) | 1.051 (1.005 to 1.100) |
| Screening tool used | | | | |
| Kryptor | 0.0000 | 1.0000 | 0.0000 | 1.000 |
| Delfia Xpress | −0.1191 (−0.1240 to −0.1143) | 0.7601 (0.7517 to 0.7686) | 0.0054 (0.0025 to 0.0082) | 1.012 (1.006 to 1.019) |
| Center | | | | |
| A | 0.0000 | 1.0000 | 0.0000 | 1.000 |
| B | 0.0065 (0.0039 to 0.0090) | 1.0150 (1.0091 to 1.0210) | 0.0054 (0.0025 to 0.0082) | 1.012 (1.006 to 1.019) |
| C | −0.0483 (−0.0542 to −0.0423) | 0.8948 (0.8827 to 0.9072) | 0.0351 (0.0284 to 0.0418) | 1.084 (1.068 to 1.101) |
| D | −0.0803 (−0.0861 to −0.0744) | 0.8312 (0.8201 to 0.8425) | −0.0358 (−0.0423 to −0.0292) | 0.921 (0.907 to 0.935) |

IVF, *in-vitro* fertilization.

**Table 3** Estimated median multiples of the median (MoM) values according to gestational age for free β-human chorionic gonadotropin (β-hCG) and pregnancy-associated plasma protein-A (PAPP-A) in trisomy 21 fetuses

| | | *Free β-hCG* | *PAPP-A* |
|---|---|---|---|
| *Week* | n | *MoM (95% CI)* | *MoM (95% CI)* |
| 8 | 26 | 1.2408 (0.9793–1.5723) | 0.3967 (0.3168–0.4968) |
| 9 | 46 | 1.3433 (1.1243–1.6050) | 0.3476 (0.2935–0.4117) |
| 10 | 41 | 1.3303 (1.1017–1.6063) | 0.3699 (0.3092–0.4425) |
| 11 | 116 | 1.8122 (1.6201–2.0272) | 0.4145 (0.3726–0.4611) |
| 12 | 417 | 1.9327 (1.8218–2.0504) | 0.5057 (0.4781–0.5349) |
| 13 | 231 | 2.0633 (1.9058–2.2339) | 0.6158 (0.5710–0.6640) |
| 14 | 9 | 2.1992 (1.4706–3.2886) | 0.6449 (0.4399–0.9453) |

at 8–13 weeks for maternal characteristics, method of conception and for different centers and machines show an adequate model fit (Figure 2).

## Screening performance by maternal age, serum biochemistry and nuchal translucency

In the trisomy 21 pregnancies, compared to unaffected pregnancies, free β-hCG was increased and PAPP-A was decreased (Figure 3). Estimated weekly medians with 95% confidence intervals are presented in Table 3. The quadratic term for log MoM PAPP-A was highly significant ($P = 0.0006$) while that for log MoM free β-hCG was not significant ($P = 0.4$). The standard deviations of log MoM values for PAPP-A and free β-hCG and the correlations between them are shown in Table 4.

The fitted regression models for trisomy 21 were:

$$\text{Log}_{10}\ \text{free β-hCG MoM} = 0.2233838 + (0.005761 \times (\text{GA} - 77))$$

and

$$\text{Log}_{10}\ \text{PAPP-A MoM} = -0.419464 + (0.00776061 \times (\text{GA} - 77)) + (0.000271947 \times (\text{GA} - 77)^2),$$

where GA is gestational age in days.

## Screening performance by maternal age and serum biochemistry

Standardized and model-based detection rates for trisomy 21 at fixed false-positive rates of 3 and 5% in screening by maternal age and measurements of both free β-hCG and PAPP-A at 8, 9, 10, 11, 12 and 13 weeks are shown in Figure 4.

Model-based estimates of the performance of screening for trisomy 21 by a combination of maternal age, fetal NT at 12 weeks and measurements of both free β-hCG and PAPP-A at 8, 9, 10, 11, 12 and 13 weeks as well as

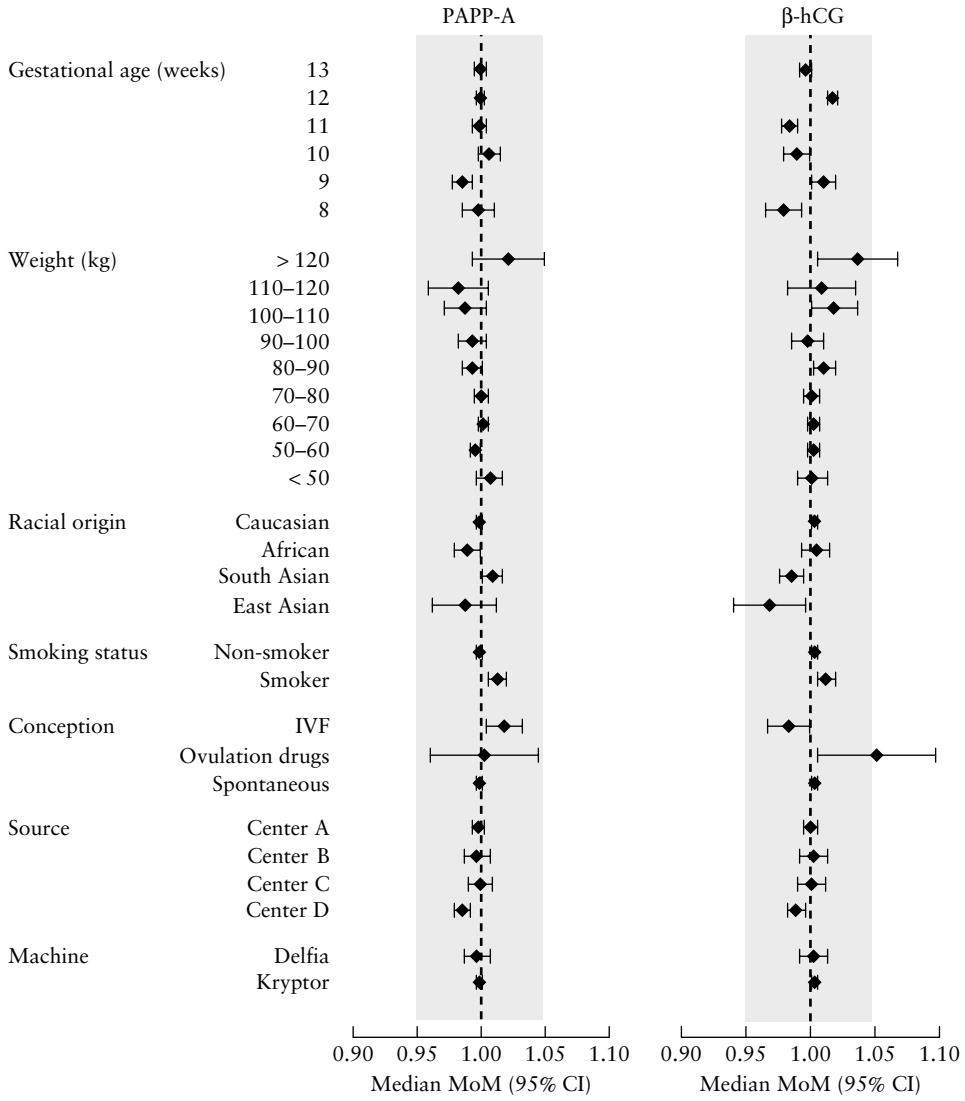


**Figure 2** Median multiples of the median (MoM) with 95% CIs diagnostics for pregnancy-associated plasma protein-A (PAPP-A) and free β-human chorionic gonadotropin (β-hCG) in chromosomally normal pregnancies. IVF, *in-vitro* fertilization.

measurement of free β-hCG and PAPP-A separately at 8, 9, 10, 11, 12 and 13 weeks are shown in Table 5 and illustrated in Figure 5.

## DISCUSSION

The findings of this large multicenter study demonstrate the gestational age-related differences in serum free β-hCG and PAPP-A between trisomy 21 and unaffected pregnancies and provide estimates of the performance of first-trimester screening utilizing different strategies in terms of the selected gestation for measurement of free β-hCG and PAPP-A. The key contributions of the paper are firstly, improved regression models for standardizing PAPP-A and free β-hCG for gestation and weight that fit well across the range of 7 to 14 weeks' gestation; secondly, new regression models for PAPP-A and free β-hCG MoM values in trisomy 21 pregnancies; and thirdly, demonstration that in combined testing PAPP-A should not be measured before 9 weeks because evidence suggests that the separation in log MoM PAPP-A between trisomy 21 and unaffected pregnancies is maximum at 9–10 weeks and narrows before this gestational age, and the spread in log MoM values for PAPP-A increases very early in gestation.

In unaffected pregnancies the maternal serum concentration of PAPP-A increases with gestation between 7 and 14 weeks. Between 8 and 9 weeks, PAPP-A concentrations double and as gestation increases the rate of increase drops to around 50% between weeks 12 and 13. Free β-hCG increases to a maximum at around 9 weeks and decreases thereafter.

The measured serum concentrations of these placental products were affected by maternal characteristics, including racial origin, weight and method of conception as well as the machine and reagents used for the analysis. Consequently, we used multiple regression analysis to estimate the effects of these

 

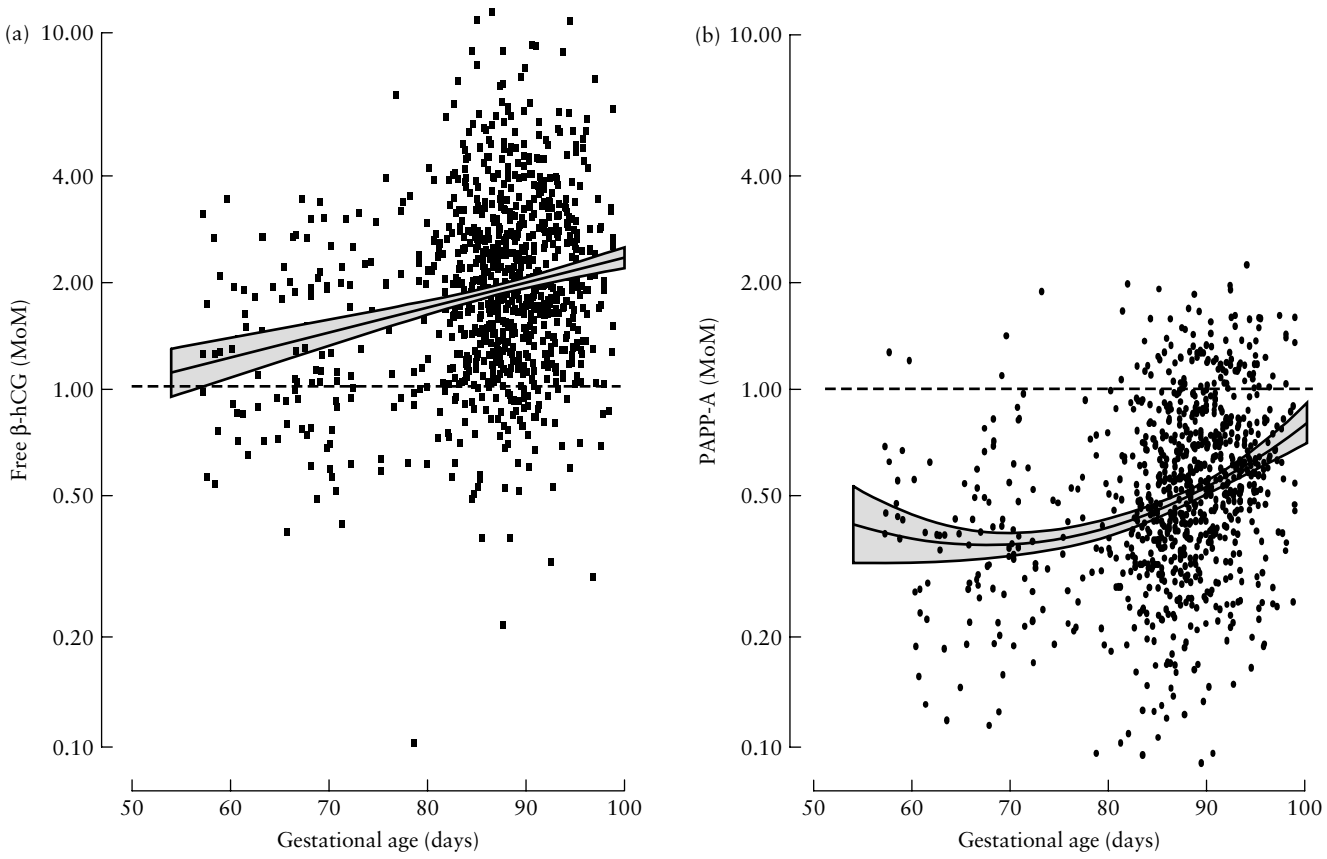


**Figure 3** Distribution of free β-human chorionic gonadotropin (β-hCG) (a) and pregnancy-associated plasma protein-A (PAPP-A) (b) multiples of the median (MoM) according to gestational age in trisomy 21 fetuses with their regression lines and 95% CIs.

**Table 4** Estimated distributional parameters for log multiples of the median of pregnancy-associated plasma protein-A (PAPP-A) and free β-human chorionic gonadotropin (β-hCG) in trisomy 21 and unaffected pregnancies

| *GA when tested* | *Standard deviation* | | *Correlation coefficient* |
|---|---|---|---|
| | *PAPP-A* | *Free β-hCG* | |
| Trisomy 21 pregnancies | | | |
| 8 weeks | 0.2382 | 0.2197 | 0.1548 |
| 9 weeks | 0.2490 | 0.2329 | 0.1535 |
| 10 weeks | 0.2278 | 0.2631 | 0.1900 |
| 11 weeks | 0.2528 | 0.2447 | −0.0973 |
| 12 weeks | 0.2392 | 0.2635 | 0.1234 |
| 13–14 weeks | 0.2326 | 0.2464 | 0.1500 |
| All | 0.2388 | 0.2537 | 0.1054 |
| Unaffected pregnancies | | | |
| 7–8 weeks | 0.2456 | 0.2354 | 0.3788 |
| 9 weeks | 0.2393 | 0.2366 | 0.3036 |
| 10 weeks | 0.2349 | 0.2466 | 0.2388 |
| 11 weeks | 0.2324 | 0.2584 | 0.2188 |
| 12 weeks | 0.2221 | 0.2544 | 0.2153 |
| 13–14 weeks | 0.2158 | 0.2542 | 0.2261 |
| All | 0.2251 | 0.2527 | 0.2312 |

GA, gestational age.

maternal variables in defining MoMs before comparing affected and unaffected pregnancies. The effects were similar to our previously published estimates[4]. Another important finding highlighted by our data is that, despite corrections for maternal characteristics and reagents used for the analysis, there are variations in the median MoMs of free β-hCG and PAPP-A between different centers. Consequently, provision of accurate patient-specific risks requires local adjustments in each laboratory to achieve a median MoM value of 1.0 MoM.

In trisomy 21 pregnancies the mean log MoM free β-hCG increased linearly with gestation between 8 and 13 weeks, as shown previously by Spencer *et al.*[2,3]. In contrast, the relationship between log MoM PAPP-A and gestation was fitted by a quadratic equation such that the maximum separation between trisomy 21 and unaffected pregnancies occurs at 9–10 weeks. As a consequence, the performance of first-trimester biochemical screening was best at 9–10 weeks rather than 7–8 or 11–14 weeks. This is consistent with the results of a meta-analysis of trisomy 21 pregnancies at 6–20 weeks that reported a quadratic relationship for MoM PAPP-A with a maximum separation between trisomy 21 and unaffected pregnancies at 8–9 weeks[10].

Effective first-trimester screening for trisomy 21 is provided by combining maternal age and serum biochemistry with ultrasound examination for the measurement of fetal NT thickness. However, the aim of the first-trimester scan is not just to screen for trisomy 21 but also to diagnose an increasing number of fetal malformations, and in this respect the ability

 

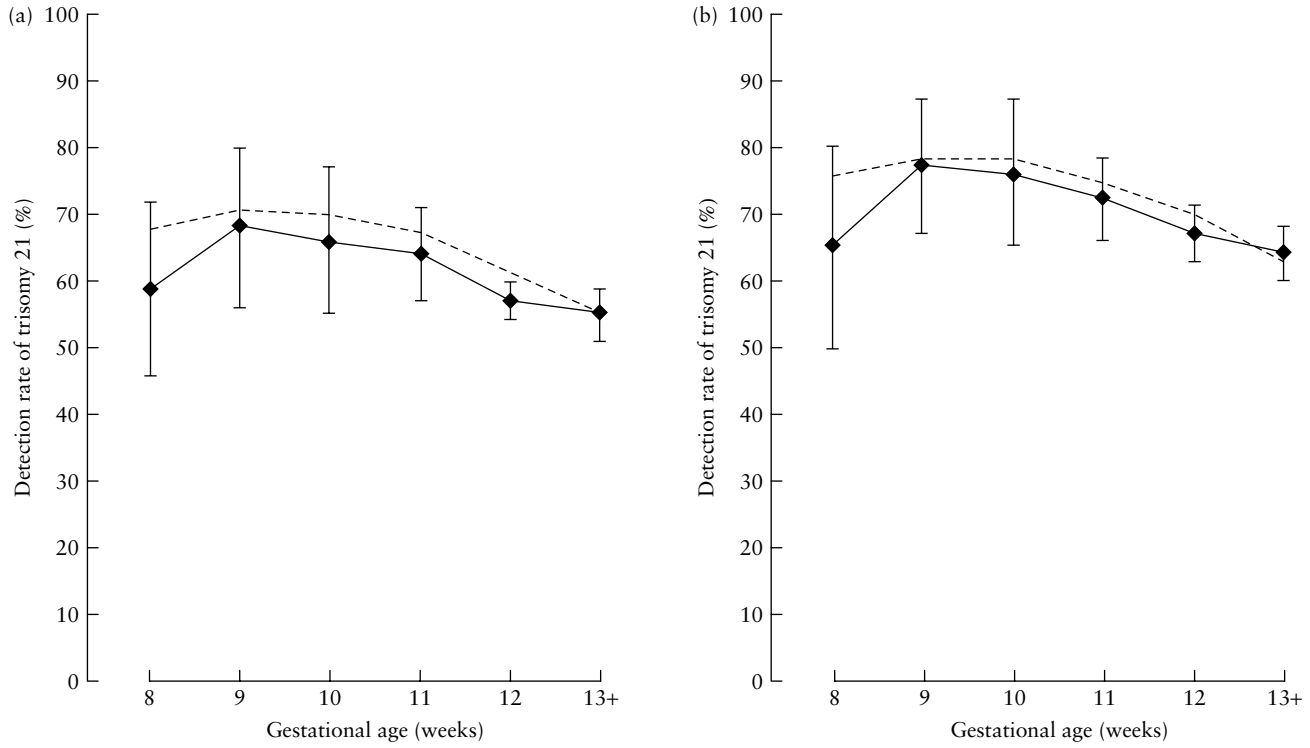


**Figure 4** Detection rates of trisomy 21 at fixed false-positive rates of 3% (a) and 5% (b) in screening by maternal age and measurements of both free β-human chorionic gonadotropin and pregnancy-associated plasma protein-A at 8, 9, 10, 11, 12 and 13 weeks' gestation. - - - - , model-based rate; ———, empirical standardized rate (with 95% CIs).

**Table 5** Model-based estimates of detection rates (%) for trisomy 21 at a fixed false-positive rate of 3% (2 and 5% given in parentheses) in combined screening by maternal age, fetal nuchal translucency thickness at 12 weeks' gestation and measurements of free pregnancy-associated plasma protein-A (PAPP-A) and free β-human chorionic gonadotropin (β-hCG) at different gestational ages

| *Gestational age at PAPP-A test (weeks)* | *Gestational age at free β-hCG test (weeks)* | | | | | |
|---|---|---|---|---|---|---|
| | *8* | *9* | *10* | *11* | *12* | *13* |
| 8 | 89 (85, 92) | 89 (87, 92) | 90 (87, 93) | 91 (89, 94) | 92 (90, 95) | 93 (91, 95) |
| 9 | 89 (86, 92) | 90 (87, 93) | 90 (88, 94) | 91 (89, 94) | 92 (90, 95) | 94 (92, 96) |
| 10 | 88 (85, 91) | 89 (85, 92) | 89 (87, 92) | 91 (88, 93) | 91 (89, 94) | 92 (90, 95) |
| 11 | 85 (82, 89) | 86 (82, 90) | 87 (84, 91) | 88 (85, 92) | 90 (87, 92) | 91 (88, 94) |
| 12 | 81 (77, 86) | 82 (78, 86) | 83 (80, 88) | 85 (81, 88) | 86 (83, 90) | 88 (84, 91) |
| 13 | 76 (71, 81) | 77 (73, 83) | 79 (74, 84) | 80 (77, 85) | 82 (78, 86) | 84 (80, 88) |

to visualize fetal anatomy is best at 12 weeks[11]. One option in screening for trisomy 21 is to perform biochemical testing and the ultrasound scan in the same visit at 12 weeks, with estimated detection rates of 86 and 90% at false positive rates of 3 and 5%, respectively.

An alternative strategy for first-trimester combined screening is for biochemical testing and ultrasound scanning to be carried out in two separate visits, with the first done at 9 weeks and the second at 12 weeks. For false-positive rates of 3 and 5% the estimated detection rates would be 90 and 93%, respectively. The improved performance of such two-stage first-trimester screening has already been reported from two previous prospective studies[5,12].

A third option would be to perform the scan at 12 weeks and optimize the performance of biochemical testing by measuring PAPP-A at 9 weeks and free β-hCG at the time of the scan at 12 weeks or even later, with estimated detection rates of 92 and 95% at respective false-positive rates of 3 and 5%.

This study has demonstrated the potential advantage, in terms of detection and false-positive rates, of offering biochemical testing and the ultrasound scan at different gestations within the first trimester. The cost and patient acceptability of the alternative policies of such first-trimester testing will depend on the existing infrastructure of antenatal care, which will vary between different countries and centers.

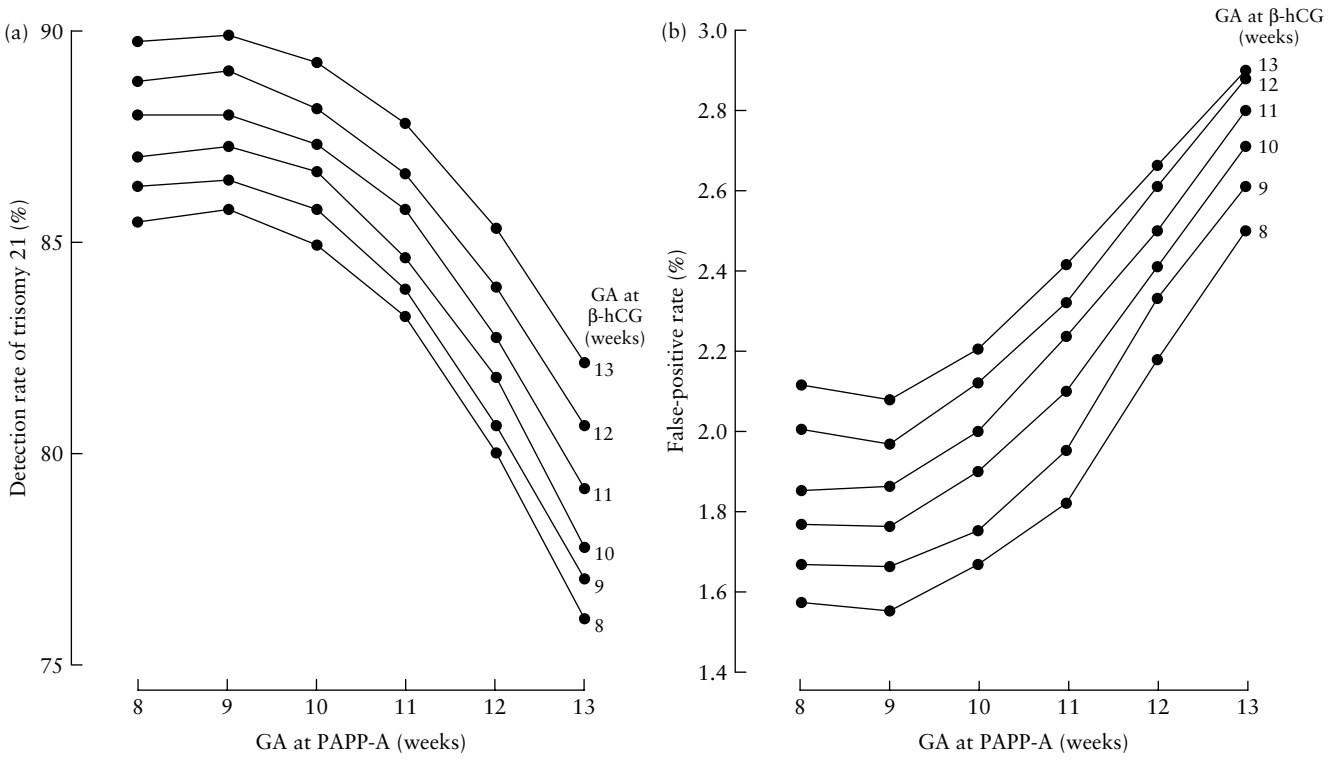


**Figure 5** Model-based estimates of detection rate for trisomy 21 (a) and false-positive rate (b) at a fixed risk cut-off of 1 in 100 in combined screening by maternal age, fetal nuchal translucency thickness at 12 weeks' gestation and measurements of β-human chorionic gonadotropin (β-hCG) and pregnancy-associated plasma protein-A (PAPP-A) at different gestational ages (GA) of 8, 9, 10, 11, 12 and 13 weeks.

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