First- and Second-trimester Down Syndrome Screening: Current Strategies and Clinical Guidelines

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SUMMARY

Down syndrome (DS) is the most common human disease caused by a structural chromosome defect. The original screening test for DS was maternal age or a history of a previously affected infant. Maternal serum screening has been incorporated into routine prenatal checkup in Taiwan since 1994. We used free β-human chorionic gonadotropin and α-fetoprotein (double test) as the serum markers, and this was carried out between the 15th to 20th week of gestation. The overall detection rate was 56% and was compatible with studies of Caucasian populations. The impact of double tests in Taiwan has shown itself by a dramatic lowering of the rate of DS live birth from 0.63 before screening to 0.16 per 1,000 live births at present. However, because of its relatively low detection rate and poor cost-effectiveness, the double test is not justified as a routine screening tool currently. First-trimester combined test is now becoming more widely available and provides increased sensitivity when detecting DS; it has a detection rate of approximately 85% with a false-positive rate of 5%. Nuchal translucency measurement requires ongoing quality control and sufficient certificated obstetricians; therefore, first-trimester ultrasound is limited only in designated centers. The quadruple test, having comparable detection rate, should be considered for incorporation into second-trimester screening in Taiwan in the near future. Other screening approaches and combinations have also been utilized in the Western countries. In this review, we outline the various options with respect to DS screening and hope that this will provide practical information for physicians offering such screenings. [*Taiwan J Obstet Gynecol* 2008;47(2):157–162]

Key Words: Down syndrome, first trimester, prenatal screening, second trimester

Introduction

Down syndrome (DS) is the most common human disease caused by a structural chromosome defect, with an occurrence rate of about 1 in 700 [1,2]. The original screening test for DS was maternal age or a history of a previously affected infant. Women who are 35 years or older account for approximately 30% of DS, which can be diagnosed mostly by amniocentesis [3,4]. Maternal serum screening has been incorporated into the routine prenatal checkup in Taiwan since 1994 [5–8]. Free β-human chorionic gonadotropin (free β-hCG) and α-fetoprotein (AFP) are used as serum markers between the 15th and 20th week of gestation. The impact of second-trimester screening has shown itself by a dramatic lowering of the rate of DS live birth from 0.63 to 0.16 per 1,000 live births [7]. Nuchal translucency (NT) measurement as an approach to DS screening was firstly reported by Snijders et al [9] in 1998. Furthermore, the

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first-trimester combined test, which includes the serum markers free β-hCG and pregnancy-associated plasma protein-A (PAPP-A), has reached a detection rate of nearly 90% [10]. There are many combinations of first- and second-trimester serum and sonographic screening tests available at present. The American College of Obstetricians and Gynecologists (ACOG) suggests that first-trimester combined test should be routine for general population screening [11] and second-trimester screening should be reserved for when a certificated sonographer is not available. This article describes the evolutional changes in DS screening strategies in Taiwan over the last few years and possible clinical guidelines for the near future.

Second-trimester Screening

Screening for DS by maternal age started 30 years ago when amniocentesis was offered only to older women (those ≥ 35 years old). Second-trimester screening, generally offered between 14th and 20th week of gestation, traditionally consists of some combination of maternal serum analysis and maternal age. In the 1980s, it was discovered that low maternal serum AFP in the second trimester was associated with an increased risk of DS [12–14]. Subsequently, the association between elevated serum hCG and DS led to the development of the "double test". The double test was introduced into routine prenatal examinations in Taiwan starting in 1994 [6]. Tables 1 and 2 show the results for the double test across the three largest studies carried out in Taiwan. The detection rate in these studies ranged from 57% to 63% with a false-positive rate of between 5.3% and 6.5%; these results are compatible with those obtained from Caucasian population studies [6–8,15]. Later, a third marker for DS, unconjugated estriol, was found to be lower in affected pregnancies [16]. This led to the "triple test", which is commonly used in many other countries [17]. The overall detection rate using the triple test increased to 69% with the same false-positive rate [11]. Recently, inhibin A has been added to form the "quadruple test"; this has significantly improved screening performance, giving detection rates that reach 81% with a 5% false-positive rate [10, 15,18]. The quadruple test is currently the most popular second-trimester screening test in the USA. Wald et al [19] suggested that the double test is no longer justified as a routine screening tool for DS on the basis of the relatively low detection rates and poor cost-effectiveness. In Taiwan, we believe that the quadruple test should be considered for incorporation into second-trimester screening.

In terms of routine screening in Taiwan, we need to focus on young mothers. In Taiwan, women with an advanced age almost always choose genetic diagnosis [20]. The low cost of genetic diagnosis in Taiwan and a poor support system for intellectually disabled children have resulted in Taiwanese women preferring to receive amniocentesis rather than risk a DS baby, despite the fetal loss rate with amniocentesis of nearly 1 in 300. However, if the second-trimester serum test is offered to the young group only, the overall detection rate is dramatically decreased. From our unpublished data, the detection rate of the double test among a group of women with advanced maternal age was found to be only 45% with a false-positive rate of 3.0%; this study was carried out over 7 years from 1999 to 2005 at our institute, the results of which should be compared with the 46% detection rate from the study by Wald et al [15]. When the results of three major studies in Taiwan are correlated, the average detection rate among women of advanced maternal age is 49% (Tables 1 and 2).

In Taiwan, over 80% of pregnant women have their babies delivered in a local hospital or a private clinic, and therefore, it would be very difficult to train all obstetricians as qualified sonographers to fully cover all prenatal services. Furthermore, AFP in the second trimester could still play a role in detecting neural tube

Table 1. First- and second-trimester Down syndrome screening tests and detection rates (with a 5% false-positive rate)

	Detection rate (%)	References
First-trimester NT measurement alone	70	22
First-trimester combined test	82–87	10, 11
Second-trimester double test	57–63	6, 8, 15
Second-trimester triple test	69	15
Second-trimester quadruple test	81	10, 11
Integrated test	94	10, 11
Stepwise sequential test	95	10
Contingent sequential test	88–94	39

NT = nuchal translucency.

Table 2. Studies of the second-trimester double test in Taiwan

Total number of DS cases	Number of DS cases detected	AMA	DR/FPR in all cases	DR/FPR in maternal age group 15–34 years old	DR/FPR in maternal age group > 34 years old	Authors
23	13	12	57% (13/23)/5.3% (93/1,748)	45% (5/11)/4.1% (60/1,458)	67% (8/12)/11.4% (33/290)	Hsu et al. [6]
16	10	2	63% (10/16)/6.5% (1,154/17,742)	57% (8/14)/5.1% (880/16,549)	100% (2/2)/23% (274/1,193)	Jou et al. [8]
22	11	2	50% (11/22)/3.2% (560/17,486)	45% (9/20)/3.0% (477/15,912)	100% (2/2)/5.3% (83/1,574)	Shaw et al* (unpublished data, 1999–2005)
61	34	16	56% (34/61)/4.9% (1,807/36,976)	49% (22/45)/4.2% (1,417/33,919)	75% (12/16)/12.8% (390/3,057)	Hsu et al [6], Jou et al [8], Shaw et al*
77	–	–	61%/5.6%	46%/4.0%	86%/2.4%	Wald et al [15]

**Oral presentation on March 15, 2008. Annual meeting of the Taiwan Association of Obstetrics and Gynecology. DS = Down syndrome; AMA = advanced maternal age; DR = detection rate; FPR = false-positive rate.*

defects [11]. Although the detection rate is higher using the first-trimester combined test, second-trimester testing would seem to be more convenient and more reliable for most obstetrics clinics in Taiwan.

First-trimester Screening

First-trimester screening is typically conducted between the 11th and 14th week of gestation. At this time, NT is a powerful sonographic marker for DS, and free β-hCG and PAPP-A are the discriminatory serum factors [9,21,22]. These three markers are used to calculate the likelihood ratio, which is used to modify the woman's age-related risk and thus determine the individual risk of fetal DS [23]. The performance of NT screening in terms of success varies among studies, with the detection rate ranging from 64% to 70% with a 5% false-positive rate depending on the study [10]. Furthermore, an increased thickness of the NT may also be associated with other chromosomal abnormalities [24–27]. During the early 1990s, several studies reported the association between DS and a low level of PAPP-A during the first trimester. A similar association was found between DS and a high level of hCG during the first trimester [28,29]. A combination of NT measurement with the above two serum biochemical markers in the first trimester comprises the "combined test" [23]. The detection rate for this test is between 82% and 87% with a false-positive rate of 5%, which is even better than quadruple test in the second trimester [10]. The advantage of the combined test is the availability of the results in the late first trimester, which allows karyotyping by chorionic villus sampling and early surgical termination of pregnancy when this is indicated. The ACOG's 2007 clinical guideline concluded that the first-trimester combined test is an effective screening test for DS for the general population (level A evidence) [11]. The first-trimester combined test was introduced at major medical centers in Taiwan in the beginning of 2006 and was based on the Fetal Medicine Foundation (FMF) guideline. At present, there are 34 FMF certified sonographers in our country. The Taiwan Society of Perinatology announced that the first-trimester combined test ought to be incorporated into routine care officially. One study showed that the first-trimester combined test was the most cost-effective screening tool [30]. We think that DS screening will move towards first-trimester testing as a gold standard in the near future and that this revolutionary change will occur in the next 10 years. However, the relatively high abortion rate with chorionic villus sampling and the possibility

of operator error with sonography should be considered by clinical doctors. In addition, patients need to be informed on these facts too.

The FMF at the King's College Hospital in London developed the first clinical training program for sonographers that taught the appropriate technique for measuring NT. Software for determining the risk assessment of DS using this NT measurement is made available to those completing the training along with an ongoing quality assurance system. The results suggest that this program of training and quality assurance yields consistently high-quality measurements [31].

Other Soft Markers in the First Trimester

Nasal bone

Cicero et al [32] published the first large prospective trial involving nasal bone assessment in a high-risk population undergoing chorionic villus sampling to assess for chromosomal abnormalities. They found that absence of the nasal bone during first-trimester scanning was associated with DS. The nasal bone was absent in 43 of 59 (73%) DS fetuses and in only three out of 603 (0.5%) euploid fetuses. Based on the high likelihood ratio for DS with an absent nasal bone and a similarly low negative likelihood ratio when the nasal bone was present, this study estimated that nasal bone assessment would significantly improve the performance of first-trimester ultrasound testing for DS.

Ductus venosus

The ductus venosus is a blood vessel present during embryonic and fetal life that originates from the umbilical vein and empties into the inferior vena cava just proximal to its entry into the right atrium. The normal Doppler waveform of the ductus venosus is pulsatile and should always demonstrate forward flow. However, reversed flow has been associated with both aneuploidy and congenital heart disease [33].

Tricuspid regurgitation

In 2003, Huggon et al [34] reported the relationship between tricuspid regurgitation and aneuploidy in women referred for early fetal echocardiography. Abnormal karyotypes were found in 83% of fetuses with tricuspid regurgitation. To date, only a few studies linking tricuspid regurgitation with DS have been published. Further investigations in this area may prove productive. Like ductus venosus, measuring the standard tricuspid regurgitation needs to be performed by a well-trained sonographer.

First- Plus Second-trimester Screening

The integrated test

The integrated test combines first-trimester NT measurement and serum PAPP-A levels with second-trimester AFP, β-hCG, unconjugated estriol and inhibin A (the quadruple test) [15]. In an initial report, the estimated detection rate for the integrated test was 94%. The major advantage of this test is its high detection rate, which implies that fewer women will need to undergo invasive testing with the inherent risk of miscarriage using the integrated test; and equally importantly, fewer women will be made anxious about their pregnancy [35]. The integrated test has been challenged ethically, since the integration of first- and second-trimester markers in a single test could pose a problem with respect to the withholding of first-trimester result, thus denying the mother the possible advantages of an earlier pregnancy termination [36]. Another major disadvantage is the relatively high cost. As a population-based screening tool, the cost would be very high if all women received both first- and second-trimester testing.

The sequential test

Sequential testing involves the performance of both first- and second-trimester testing, but with the immediate disclosure of first-trimester results for use in clinical management. There are three approaches to such sequential risk management: (1) independent, (2) stepwise, and (3) contingent.

1. Independent sequential testing involves the independent interpretation of first-trimester combined test and second-trimester serum test. The first-trimester result is given to the patient for clinical decision-making. The second-trimester test is interpreted without taking into account the first-trimester results, i.e. maternal age is used as the *a priori* risk for the second-trimester testing. Although the sensitivity is high, this is the least efficient risk assessment strategy, because the test's additive false-positive rate is unacceptably high [10,37].
2. Stepwise sequential testing suggests an early invasive procedure if the first-trimester result is above a specific cut-off. If the first-trimester risk assessment result is below this cut-off, then the patient is offered second-trimester testing, with the final risk being determined using all the markers. A detection rate of 95% with a 4.9% false-positive rate has been shown [10]. The advantage of this approach is sensitivity and a false-positive rate approaching that obtained using integrated risk assessment, but with the option that the early

results are available in the first trimester for the highest-risk patients.

3. Contingent sequential testing begins with the performance of first-trimester risk assessment. On the basis of these results, women are grouped into one of three risk categories: high-risk, intermediate-risk, and low-risk. The cut-off points of the groups and their specific risks vary, depending upon how these groups are defined [38]. For contingent sequential risk assessment to be successful, careful determination of the risk cut-offs is required. The first-trimester cut-off must identify a significant proportion of DS pregnancies with only a small number of false-positives. In one study, the detection rate was 88–94% with a 5% false-positive rate [39]. Of all the potential screening protocols, contingent sequential screening seems to have the most advantages. It should achieve a high detection rate with a very low false-positive rate. Most importantly, the majority of women would have their screening completed in the first trimester, which will substantially reduce patient anxiety and increase test satisfaction. Patients would benefit from both first- and second-trimester screening, yet the obstetricians would not have to hold back information or wait until the second trimester to disclose the test results. Thus, this protocol is highly likely to increase physician satisfaction as well.

Multiple Pregnancies

Multiple pregnancy is another topic that is associated with serum screening. A recent consensus is that first-trimester NT measurement is superior to second-trimester screening [40]. All monochorionic pregnancies are monozygotic; however, not all dichorionic pregnancies are dizygotic. The risk of DS for each fetus is independent in dizygotic twin pregnancies. This indicates that for dichorionic twin pregnancies, the pregnancy specific risk needs to be calculated by summing the individual risk estimates for each fetus [41]. On the contrary, the risk would be based on the average of likelihood ratios derived from nuchal measurements of both twins in monochorionic pregnancies. Therefore, diagnosis of chorionicity should be the first step in ultrasound evaluation of twins during the first trimester.

Current Strategies and Conclusion

We had offered the second-trimester double test for over 14 years and then started to set up a first-trimester platform 2 years ago. Pregnant women in Taiwan now have a choice in terms of DS screening, namely first- or second-trimester screening. The cost of first-trimester DS screening is about twice that of second-trimester DS screening. However, the detection rate of double test is only about 50%. Doctors in obstetrics across Taiwan should offer a better screening tool in the second trimester, namely triple or quadruple test, and replace the double test. Clinical physicians with certification for first-trimester nuchal scanning are able to provide first-trimester screening during early pregnancy or in cases of high-risk pregnancy with an obvious family history. Nonetheless, for those doctors without certification or those in rural areas, a more effective second-trimester screening is essential. The detection rate of the quadruple test is comparable with that of the first-trimester combined test. If the first-trimester combined test and the second-trimester quadruple test were popularly available in Taiwan and an official consensus announced by the Taiwan Society of Perinatology, the live birth rate of DS babies in Taiwan would decrease even further to an all-time low very quickly.

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