AN "IN HOUSE" SOFTWARE FOR CALCULATING LIKELIHOOD RATIO OF TRISOMY 21, 18 AND 13 BASING ON MATERNAL AGE, GESTATION, NUCHAL TRANSLUCENCY AND NASAL BONE

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Abstract

Objectives: Using Excel program and its tools to design an "in house" software to calculate the risk of trisomy 21, 18, 13. **Method:** Basing on the results in the researches to get the necessary mathematical formulae for risk calculation basing on the maternal and gestational age, previous trisomic child, nuchal translucency, nasal bone. The risks calculated by "in house" software of 270 pregnant women are compared to the risks calculated by software of Fetal Medicine Foundation (FMF). **Results:** An"in house" software has been easily designed for calculating, printing and saving. There was the almost perfect agreement between 2 softwares for all cases in groups. **Conclusion:** The "in house" software can be used by Vietnamese sonographers after training in prenatal scan to calculate the risk of trisomy 21, 18 and 13 and supply advice to pregnant women.

1. INTRODUCTION

Down's syndrome is the most common congenital cause of severe mental retardation, with an incidence at birth of about 1-3 per 1000. In 2006, the Centers for Disease Control and Prevention estimated the rate as one per 733 live births in the United States [1]. Approximately 95% of these are trisomy 21. Down syndrome occurs in all ethnic groups and among all economic classes.

The first method of screening for trisomy 21, introduced in the early 1970s, was based on the association with advanced maternal age [2]. In the late 1980s, a new method of screening was based on not only maternal age but also the concentration of various fetoplacental products in the maternal circulation at 16 weeks of gestation, the maternal serum concentrations of alpha-fetoprotein (AFP), uconjugated estriol (uE3), human chorionic gonadotropin (hCG) (total and free-b) and inhibin-A. This method of screening is more effective than maternal age alone and it can identify about 50–70% of the fetuses with trisomy 21 [3,4].

In the 1990s, screening by a combination of maternal age, fetal NT and maternal serum biochemistry (free b-hCG and PAPP-A) in the first-trimester identify about 85–90% of affected fetuses [5].

In 2000s, screening by a combination of maternal age maternal age, fetal NT, maternal serum biochemistry and nasal bone not visible by ultrasound at $11–13^{+6}$ weeks can increase the detection rate of the first trimester scan and serum biochemistry to more than 95% [6,7] (table 1).

Table 1. The comparison of the detection rates (DR), for a false positive rate of 5%, of different methods of screening for trisomy 21

Method of screening	DR(%)
Maternal age (MA)	30
MA and maternal serum biochemistry at 15–18 weeks	50–70
MA and fetal nuchal translucency (NT) at 11–13+6 wks	70–80
MA and fetal NT and maternal serum free b-hCG and PAPP-A at 11–13+6 wks	85–90
MA and fetal NT and fetal nasal bone (NB) at 11–13+6 wks	90
MA and fetal NT and NB and maternal serum free b-hCG and PAPP-A at 11–13+6 wk	95

hCG human chorionic gonadotropin, PAPP-A: pregnancy-associated plasma protein A

In condition, the ultrasound day by day becomes more popular in Vietnam, a lot of training courses for sonographers in the field of prenatal scan, especially nuchal ranslucency scan have been organized. The need of a software for calculating the risk of trisomy 21, 18, 13 basing on maternal age, gestational age, previous trisomy child, nuchal translucency, nasal bone written by Vietnamese language, easy to use is necessary for sonographers after training in the field of screening these trisomies.

Basing on the articles, it has been announced that we have used Excel program to design an "in house" software for calculating the risk of trisomy 21, 18 and 13.

2. PRENATAL SCREENING TRISOMY 21, 18 AND 13 IN VIETNAM

In South Vietnam

In 2007, there was the first report of prenatal screening and diagnosis in second trimester for trisomy 21 with triple test[1] using kits of T21 Kit – Gamma (Belgium) and patient specific risks were calculated by the software T21 Gamma (Belgium). The research was carried out from 2004 – 2005 in Tu Du Hospital and University of Medicine and Pharmacy in HCM city, South Vietnam on 2435 pregnant women [8].

In the same year, this group of authors established the median values for maternal serum markers in triple test by ELISA technique for T21 Kit-Gamma (Belgium) using on the semiautomatic equipment [9].

In 2007, there was the first report from 2004 – 2006 [10] of using the FISH technique in prenatal diagnosis for some chromosomal aneuploidies in the laboratory of cytogenetics of University of Medicine and Pharmacy in HCM city on 1302 samples of amniotic fluids.

In 2009, there was the second report of prenatal screening in second trimester for Down Syndrome by triple test but tested on a better system: the automatical Immulite 2000 system and Prisca software, on 6193 pregnant women from 2007 – 2008 in South Vietnam [11].

In the same year, there was the first report of prenatal screening by combined test[2] on 2674 pregnant women with the gestational age from 11 – 13^{+6} weeks from 2007 - 2008. The Prisca software was used to calculate the patient specific risk basing on the measurement of

[1]*Second-trimester test based on the measurement of AFP, uE3, and hCG (either total hCG or free β-hCG) together with maternal age.*

[2]*First-trimester test based on combining nuchal translucency measurement with free β-hCG,PAPP-A and maternal age.*

nuchal translucency, the levels of PAPP-A and free β hCG in maternal serum. This research was carried out in Tu Du Hospital and University of Medicine and Pharmacy in HCM city [12].

In North Vietnam

There was not a lot information related to the researches in this field from North Vietnam

In the seminar ***"Evaluation of performance in 2008 and oriented action plan in 2009 of the hospitals participating in the project improving population quality"*** held in Hanoi in the 3rd December. In a report of The National Hospital of Obstetrics and Gynecology, Hanoi, 1377 pregnant women took part in the prenatal screening program in the first six months of the year 2008.

From 2006, there has been a report of prenatal screening for Down syndrome by triple test of the authors in Hanoi Medical School [13] but until now there is no any report related to the use of combined test in prenatal screening.

In Central Vietnam

From 2008, the triple test was done in the lab of Department of Human Genetics for screening Down syndrome in second trimester. This test couldn't be long because of the restriction of the number of pregnant women who took part in the program. The software T21 Gamma has been used for calculating the risk of trisomy 21, 18 and 13. In 2009, Hue University of Medicine and Pharmacy established the Center of Prenatal - Neonatal screening and diagnosis basing on the decision of Ministry of Health. This center is responsibility for the prenatal and neonatal screening and diagnosis for 7 provinces of Central Vietnam (Quang Binh, Quang Tri, Da Nang, Quang Nam, Quang Ngai, Binh Dinh, Gia Lai).

3. METHODOLOGY

Calculation procedure for the "in house" software

It was based on the results in the researches to get the necessary mathematical formulae for risk calculation basing on the maternal and gestational age, previous trisomy child, nuchal translucency, nasal bone.

Excel program of Microsoft Office has been used to design an "in house" software for calculating the risk basing on these formulae. Macros have been used to help users calculate the risk of trisomy 21, 18 and 13, printing and saving quickly.

Compare to another software

We compared our "in house" software (IS) with the First Trimester Screening Program version 2.3.0_11, a software of Fetal Medicine Foundation (FMF) [14] for first trimester of gestation basing on the data of pretend 270 pregnant women. They were divided into three groups. In each group, the same data would be input in 2 softwares for getting the risk of trisomy 21, 18 and 13. Using the risk cutting off for Trisomy 21 was the risk of woman at 35 years old with same gestational age and risk cutting off for Trisomy 18 and 13 was 1: 150 [15] to decide positive or negative screening results. The results of two softwares would be compared by kappa statistics [16].

The kappa measure of agreement was the ratio:

$$\kappa = \frac{\Pr(a) - \Pr(e)}{1 - \Pr(e)},$$

Where:

- Pr(a) was the relative observed agreement among 2 software

- Pr(e) was the hypothetical probability of chance agreement, using the observed data to calculate the probabilities of each observer randomly saying each category.

		Software of FMF		
		Positive Screening (+)	Negative Screening (-)	**Total**
In house software	Positive Screening (+)	a	c	a + c
	Negative Screening (-)	b	d	b + d
Total		a + b	c + d	a + b + c + d

Kappa = (Observed agreement - Chance agreement) / (1 - Chance agreement)

- Observed agreement Pr(a) = (a + d)/(a + b + c + d)
- Chance agreement Pr(e) = (a + c) * (a + b) + (b + d) * (c + d)
- Kappa = [Pr(a) – Pr(e)]/[1 – Pr(e)]

The values have been characterized by Landis and Koch [17]:

If:

- The values < 0 as indicating no agreement.
- 0 - 0.20 as slight.
- 0.21 - 0.40 as fair.
- 0.41 - 0.60 as moderate.
- 0.61 - 0.80 as substantial.
- 0.81 - 1 as almost perfect agreement.

Group 1:

- Same in date of birth, gestational age, crown rump length (CRL = 50cm)
- Group 1 was divided into 4 subgroup, each subgroup including 20 women, they were different in the values of nuchal translucency (NT), the measurement of NT changed from 1.2mm to 5mm, plus 0.2mm for each.

+ Subgroup 1a: no previous trisomy child
+ Subgroup 1b: there was previous trisomy 21 child
+ Subgroup 1c: there was previous trisomy 18 child
+ Subgroup 1d: there was previous trisomy 13 child

Group 2:

- Same in date of birth, gestational age, no previous trisomy child.
- Group 2 was divided into 8 subgroup, each subgroup including 20 women, they were different in the values of nuchal translucency (NT), the measurement of NT changed from 1.2mm to 5mm, plus 0.2mm for each.

+ Subgroup 2a: CRL is 55cm
+ Subgroup 2b: CRL is 60cm
+ Subgroup 2c: CRL is 65cm
+ Subgroup 2d: CRL is 70cm
+ Subgroup 2e: CRL is 75cm
+ Subgroup 2f: CRL is 80cm
+ Subgroup 2g: CRL is 85cm

Group 3:

- Including 50 women who were different in date of birth, gestational age, crown rump length and nuchal translucency.

4. RESULTS THE CONSTRUCTION AND MATHEMATIC BASIS OF THE "IN HOUSE" SOFTWARE

The nuchal translucency varied with gestation so the first step was to calculate the GA from fetal measurements and converted the markers to MoMs.

4.1. Calculate gestational age (GA) in days at NT measurement

GA in days was calculated by the formula for crown-rump length (CRL) in millimeters. We used the formula by Von Kaisenberg et al [18].

Gestation age (days) = 49,1115 + 0,5954 x CRL

4.2. Calculate estimated delivery date (EDD) and maternal age at term

Estimated delivery date (EDD) can be calculated from date of NT measurement (NTdate) and GA at NT (GA) in days. Using the following formula we calculated EDD:

$EDD = NT_{date} + (280 - GA_{in\ days})$

4.3. Calculate maternal age at EDD

Basing on knowing EDD, maternal age was calculated at EDD (EDDage) using the following formula:

EDDage = (EDD - DOB)/365

where DOB was maternal date of birth.

4.4. Calculate maternal age at the time of fertilization

Basing on knowing gestational age, time of screening (TOS), maternal age was calculated at the time of fertilization (TOFage) using the following formula:

TOFage = [(TOS - DOB) – GA]/365

where DOB was maternal date of birth.

4.5. Calculate maternal age-based background risk of trisomy 21

4.5.1. At time of term

Maternal age background risk (MBR) for Trisomy 21 at term was based on formulae of Hetch et al [19].

MBR=0.000631+EXP(-16.60785+0.2994 x age*)

(*) In our In-house software we used age at the time of fertilization.

4.5.2. Previous trisomy 21

Calculate a modified maternal age risk at the time of screening if there was a previous affected pregnancy with Trisomy 21 by increasing the background risk by 0.75% [20].

4.6. Maternal age and gestation-specific prevalence of trisomy 21

4.6.1. Trisomy 21

Maternal and gestational age-specific risks for trisomy 21 were calculated by multiplying the maternal age-specific prevalence in live births with the relative prevalence at a given gestation.

An adjustment according to relative prevalence (RP) used the following formula by Snijders et al [21] was used.

$Log_{10}(RP) = 0,2718 \times Log_{10}(w)^2 - 1,023 \times Log_{10}(w) + 0,9425$

$RP = 10^{Log10(RP)}$

Where w was the gestational week at NT measurement, and the risk for Trisomy 21 was calculated using the following adjustment:

Adjusted risk Trisomy 21(R21) = MBR x RP

4.6.2. Trisomy 18 and 13

4.6.2.1. Calculate the prevalence of trisomy 21 (MBR) in live births by formula of Hetch et al [2]

4.6.2.2. Calculate the relative prevalence for trisomy 18 and 13 at gestation age

Basing on the prevalence of trisomy 21, trisomy 18 and trisomy 13 by maternal age and

gestation, we calculated the linear regresstion equations for relative prevalences of trisomy 18 and 13 basing on the relative prevalence of trisomy 21 by the method of Snijder et al [22].

Relative prevalence of trisomy 18 (RP18):

RP18 = 1.229 x MBR - 1.141 with R^2 = 0.999

Relative prevalence of trisomy 13 (RP13):

RP13 = 0.369 x MBR – 0.332 with R^2 = 0.999

4.6.2.3. Estimated the prevalence at gestation age by multiple relative prevalence of trisomy 18 or 13 with maternal background risk (MBR)

Prevalence of Trisomy 18 by maternal age and gestation = RP18 x MBR

Prevalence of Trisomy 13 by maternal age and gestation = RP13 x MBR

4.6.2.4. Previous trisomy 18 or 13

Calculate a modified maternal age risk at the time of screening if there was a previous affected pregnancy with Trisomy 18 or 13 by increasing the background risk by 0.75% [3].

4.7. Nuchal translucency

4.7.1. Calculate NT multiple of median NT (MoM)

Using the following formula of Nicolaides et al [23] to calculate for log 10 of NT median of a normal fetus basing on CRL in millimeters:

$Log_{10}NT$ = – 0.3599 + 0,0127 x CRL – 0,000058 x CRL^2

NTmedian = 10 log10NT

NT (MoM) was calculated by dividing an NT measurement by the NT median:

NT(MoM) = NT/NTmedian

4.7.2. Calculate Delta NT multiple of median NT (MoM)

Spencer et al.[24] described the use of Deltas rather than MoMs for more accurate calculation of risk:

DeltaNT = NT of fetus (mm) – NT median

4.7.3. Mixture model of nuchal translucency thickness

We calculated the likelihood ratio by the two component mixture model of Wright et al. With parameter in fitted mixture model for nuchal translucency, we calculated for a pregnancy with fetal CRL and NT measurement by mm.

Trisomy 21[25]

4.7.3.1. CRL dependent component (normal pregnancies)

Step 1: Estimated mean (log_{10})

Step 2: Estimated standard deviation.

Step 3: The median NT for the CRL dependent process.

Step 4: Calculate NT_{MoM} of measured NT.

Step 5: Calculate the probability density (PD) at log (measured NT) for the fitted Guassian distribution (Gd dep).

$$PD = \left(\frac{1}{\sqrt{(2\pi)S}} \right) e^{-\frac{1}{2}Z^2}$$

$$Z = \frac{\left(Log_{10}(x) - M\right)}{S}$$

With

S: standard deviation; M: estimated mean

7.3.2. CRL independent component (normal pregnancies)

- Step 1: Estimated mean

- Step 2: Estimated standard deviation.

- Step 3: Calculate the probability density (PD) at log (measured NT) for the fitted Guassian distribution (Gd ind).

4.7.3.3. Mixture model (normal pregnancies)

According to the mixture model for unaffected pregnancies, the fitted logit of the proportion arising from the CRL -independent process was given by:

Fitted logit of the proportion (fit log) = -0.3319 – (0.03790 x CRLmm)

The fitted proportion was then given by :

Fitted proportion = 1/(1+exp(-(- fit Log)

The probability density for unaffected pregnancies was given by a weighted average of two Gaussian densities:

the CRL-independent process: (weight = fitted porportion)

the CRL-dependent process: (weight = 1 – fitted proportion).

This gived the fitted mixture model probability density of:

Pd n = (fitted log x Gd ind) +((1 – fitted propostion) × Gd dep)

4.7.3.4. CRL independent component (trisomy 21 pregnancies)

- Step 1: Estimated mean = 0.5330

- Step 2: Estimated standard deviation = 0.2093

- Step 3: Calculate the probability density (PD) at log(measured NT) for the fitted Guassian distribution (Gd ind).

$$PD = \left(\frac{1}{\sqrt{(2\pi)S}}\right) e^{-\frac{1}{2}Z^2}$$

$$Z = \frac{\left(\mathrm{Log}_{10}(x) - M\right)}{S}$$

With

S: standard deviation; M: estimated mean

7.3.5. Mixture model (trisomy 21 pregnancies)

According to the mixture model the estimated proportion of trisomy 21 pregnancies arising from the CRL-independent component was 0.9406.

This gived the fitted mixture model density of Probability density of trisomy 21 pregnancies (Pd T21):

Pd T21 = (0.9406 x Gd ind) + [(1 – 0.9406) x Gd dp)]

4.7.3.6. Likelihood ratio

The likelihood ratio of trisomy 21 to normal pregnancies was given by the probability density of trisomy 21 pregnancies (Pd T21) divided by the probability density for normal pregnancies (Pd n).

Trisomy 18 or 13

With parameter in fitted mixture model for nuchal translucency, we calculated likelihood ratio of trisomy 18 or 13 the same as the way we calculated likelihood ratio of trisomy 21.

8. Nasal bone

Likelihood Ratio for Trisomy 21

In normal fetuses the likelihood of having an absent nasal bone (%) (L an) was calculated by the formula of Cicero et al [26]:

L an (%) = (odds/1 + odds) x 100

Where odds=e^Y

and Y = Loge (odds) = -0.367 + 1.582 x (1 for Afro-Caribbean and 0 for Caucasian, Asian, Oriental or Mixed races) – 0.061 x CRL (in mm) + 0.349 x delta NT (in mm).

Similarly, in trisomy 21 fetuses:

Y = Loge (odds) = 2.275 – 0.032 x CRL (in mm) + 0.207 x delta NT (in mm).

- The positive likelihood ratio for trisomy 21 for absent nasal bone is derived by dividing the likelihood (%) in trisomy 21 by that in normal fetuses.

- The negative likelihood ratio for trisomy 21 for present nasal bone is derived by dividing (100 - the likelihood (%) in trisomy 21) by (100 - that in normal fetuses).

4.8. Calculate the combined likelihood ratio (Combined LR) for trisomy 21 from maternal age, gestation, NT and NB:

Combined LR = MBR x RP x LR NT x LR NB

Where:

MBR: Maternal age background risk of trisomy 21

RP: Relative prevalence at a given gestation of trisomy 21.

LR NT: Likelihood ratio of trisomy 21 in mixture model of nuchal translucency thickness.

LR NB: Likelihood ratio of trisomy 21 for absent or present nasal bone.

4.9. Calculate the combined likelihood ratio (Combined LR) for trisomy 18 or 13 from maternal age, gestation and NT:

Combined LR = MBR x RP x LR NT

Where:

MBR: Maternal age background risk of trisomy 21

RP: Relative prevalence at a given gestation of trisomy 18 or 13.

LR NT: Likelihood ratio of trisomy 18 or 13 in mixture model of nuchal translucency thickness.

5. COMPARE TO ANOTHER SOFTWARE

Group 1:

Subgroup 1a: No previous trisomy child

TRISOMY 21

		FMF		
		+	-	Total
IS	+	15	0	15
	-	0	5	5
	Total	15	5	20
Result				
Observed result:				1.00
Chance agreement:				0.63
Kappa:				**1.00**

TRISOMY 18

		FMF		
		+	-	Total
IS	+	8	0	8
	-	0	12	12
	Total	8	12	20
		Result		
Observed result:				1.00
Chance agreement:				0.52
Kappa:				**1.00**

TRISOMY 13

		FMF		
		+	-	Total
IS	+	5	1	6
	-	0	14	14
	Total	5	15	20
Result				
Observed result:				0.95
Chance agreement:				0.60
Kappa:				**0.88**

Subgroup 1b: There was previous trisomy 21 child

TRISOMY 21

		FMF		
		+	-	Total
IS	+	17	0	17
	-	0	5	5
	Total	17	5	22
Result				
Observed result:				1.00
Chance agreement:				0.65
Kappa:				**1.00**

TRISOMY 18

		FMF		
		+	-	Total
IS	+	8	0	8
	-	0	12	12
	Total	8	12	20
		Result		
Observed result:				1.00
Chance agreement:				0.52
Kappa:				**1.00**

TRISOMY 13

		FMF		
		+	-	Total
IS	+	4	2	6
	-	0	14	14
	Total	4	16	20
Result				
Observed result:				0.90
Chance agreement:				0.62
Kappa:				**0.74**

Subgroup 1c: There was previous trisomy 18 child

TRISOMY 21

FMF				
		+	-	Total
IS	+	15	0	15
	-	0	5	5
	Total	15	5	20
Result				
Observed result:				1.00
Chance agreement:				0.63
Kappa:				**1.00**

TRISOMY 18

FMF				
		+	-	Total
IS	+	13	0	13
	-	0	7	7
	Total	13	7	20
Result				
Observed result:				1.00
Chance agreement:				0.55
Kappa:				**1.00**

TRISOMY 13

FMF				
		+	-	Total
IS	+	4	2	6
	-	0	14	14
	Total	4	16	20
Result				
Observed result:				0.91
Chance agreement:				0.64
Kappa:				**0.74**

Subgroup 1d: There was previous trisomy 13 child

TRISOMY 21

FMF				
		+	-	Total
IS	+	15	0	15
	-	0	5	5
	Total	15	5	20
Result				
Observed result:				1.00
Chance agreement:				0.63
Kappa:				**1.00**

TRISOMY 18

FMF				
		+	-	Total
IS	+	8	0	8
	-	0	12	12
	Total	8	12	20
Result				
Observed result:				1.00
Chance agreement:				0.52
Kappa:				**1.00**

TRISOMY 13

FMF				
		+	-	Total
IS	+	15	1	16
	-	0	4	4
	Total	15	5	20
Result				
Observed result:				0.95
Chance agreement:				0.65
Kappa:				**0.86**

Comment: The risks of trisomy 13 in subgroup 1b and 1c had the substantial agreement between the 2 softwares (kappa = 0.74). All of the remainder was almost perfect agreement (kappa = 0.86 – 1.00).

Group 2:

Subgroup 1a: CRL = 55mm

Trisomy 21

FMF				
		+	-	Total
IS	+	14	0	14
	-	0	6	6
	Total	14	6	20
Result				
Observed result:				1.00
Chance agreement:				0.58
Kappa:				**1.00**

Trisomy 18

FMF				
		+	-	Total
IS	+	8	1	9
	-	0	11	11
	Total	8	12	20
Result				
Observed result:				0.95
Chance agreement:				0.51
Kappa:				**0.90**

Trisomy 13

FMF				
		+	-	Total
IS	+	6	1	7
	-	0	13	13
	Total	6	14	20
Result				
Observed result:				0.95
Chance agreement:				0.56
Kappa:				**0.89**

Subgroup 1b: CRL = 60mm

Trisomy 21

FMF				
		+	-	Total
IS	+	13	0	13
	-	0	7	7
	Total	13	7	20
Result				
Observed result:				1.00
Chance agreement:				0.55
Kappa:				**1.00**

Trisomy 18

FMF				
		+	-	Total
IS	+	9	0	9
	-	0	11	11
	Total	9	11	20
Result				
Observed result:				1.00
Chance agreement:				0.51
Kappa:				**1.00**

Trisomy 13

FMF				
		+	-	Total
IS	+	7	0	7
	-	0	13	13
	Total	7	13	20
Result				
Observed result:				1.00
Chance agreement:				0.55
Kappa:				**1.00**

Subgroup 1c: CRL = 65mm

TRISOMY 21

FMF				
		+	-	Total
IS	+	12	0	12
	-	0	8	8
	Total	12	8	20
Result				
Observed result:				1.00
Chance agreement:				0.52
Kappa:				**1.00**

TRISOMY 18

FMF				
		+	-	Total
IS	+	9	0	9
	-	0	11	11
	Total	9	11	20
Result				
Observed result:				1.00
Chance agreement:				0.51
Kappa:				**1.00**

TRISOMY 13

FMF				
		+	-	Total
IS	+	7	0	7
	-	0	13	13
	Total	7	13	20
Result				
Observed result:				1.00
Chance agreement:				0.55
Kappa:				**1.00**

Subgroup 1d: CRL = 70mm

TRISOMY 21

FMF				
		+	-	Total
IS	+	12	0	12
	-	0	8	8
	Total	12	8	20
Result				
Observed result:				1.00
Chance agreement:				0.52
Kappa:				**1.00**

TRISOMY 18

FMF				
		+	-	Total
IS	+	8	0	8
	-	0	12	12
	Total	8	12	20
Result				
Observed result:				1.00
Chance agreement:				0.52
Kappa:				**1.00**

TRISOMY 13

FMF				
		+	-	Total
IS	+	7	0	7
	-	0	13	13
	Total	7	13	20
Result				
Observed result:				1.00
Chance agreement:				0.55
Kappa:				**1.00**

Subgroup 1e: CRL = 75mm

TRISOMY 21

FMF				
		+	-	Total
IS	+	11	0	11
	-	0	9	9
	Total	11	9	20
Result				
Observed result:				1.00
Chance agreement:				0.51
Kappa:				**1.00**

TRISOMY 18

FMF				
		+	-	Total
IS	+	8	0	8
	-	0	12	12
	Total	8	12	20
Result				
Observed result:				1.00
Chance agreement:				0.52
Kappa:				**1.00**

TRISOMY 13

FMF				
		+	-	Total
IS	+	7	0	7
	-	0	13	13
	Total	7	13	20
Result				
Observed result:				1.00
Chance agreement:				0.55
Kappa:				**1.00**

Subgroup 1f: CRL = 80mm

TRISOMY 21

FMF				
		+	-	Total
IS	+	11	0	11
	-	0	9	9
	Total	11	9	20
Result				
Observed result:				1.00
Chance agreement:				0.51
Kappa:				**1.00**

TRISOMY 18

FMF				
		+	-	Total
IS	+	8	0	8
	-	0	12	12
	Total	8	12	20
Result				
Observed result:				1.00
Chance agreement:				0.52
Kappa:				**1.00**

TRISOMY 13

FMF				
		+	-	Total
IS	+	7	0	7
	-	0	13	13
	Total	7	13	20
Result				
Observed result:				1.00
Chance agreement:				0.55
Kappa:				**1.00**

Subgroup 1g: CRL = 85mm

TRISOMY 21

FMF				
		+	-	Total
IS	+	11	0	11
	-	0	9	9
	Total	11	9	20
Result				
Observed result:				1.00
Chance agreement:				0.51
Kappa:				**1.00**

TRISOMY 18

FMF				
		+	-	Total
IS	+	8	0	8
	-	0	12	12
	Total	8	12	20
Result				
Observed result:				1.00
Chance agreement:				0.52
Kappa:				**1.00**

TRISOMY 13

FMF				
		+	-	Total
IS	+	7	0	7
	-	0	13	13
	Total	7	13	20
Result				
Observed result:				1.00
Chance agreement:				0.55
Kappa:				**1.00**

Comment: There was the almost perfect agreement between the 2 softwares for all cases in group 1

Group 3:

Trisomy 21				
FMF				
		+	-	Total
IS	+	28	0	28
	-	0	22	22
	Total	28	22	50
Result				
Observed result:				1.00
Chance agreement:				0.51
Kappa:				**1.00**

Trisomy 18

FMF				
		+	-	Total
IS	+	6	0	6
	-	0	44	44
	Total	6	44	50
Result				
Observed result:				1.00
Chance agreement:				0.79
Kappa:				**1.00**

Trisomy 13

FMF				
		+	-	Total
IS	+	3	1	4
	-	0	46	46
	Total	3	47	50
Result				
Observed result:				0.98
Chance agreement:				0.87
Kappa:				**0.85**

Comment: There was the almost perfect agreement between the 2 softwares for all cases in group 3

6. DISCUSSION

Basing on the results, comparing between the 2 sofwares, there was the almost perfect agreement between the 2 softwares for all cases in all groups. In the group 3, there was the difference in the risk of trisomy 13 of only one case (FMT 1: 155 and IS 1: 132).

In the real condition of Vietnam, especially in Central Vietnam, when all district hospitals had good ultrasounds, computers with excel program (Microsoft office) and all the sonographers received the training for measurement the nuchal translucency and crown rump length but not easy to get the license for using the software of FMF freely. We thought that an "in house" software running in Excel would be a useful and economical solution for helping sonographers counsel to pregnant women when scanned in the first trimester of gestation.

Our Vietnamese "in house" software running on Excel basis was designed for calculating the risk basing on maternal age, gestational age, crown rump length, previous trisomy child, nuchal translucency and the nasal bone if the sonographers received the good training in scan skill. It was designed for easy to save all information of each patient and printing the results. It can be update easily and replace quickly if there is any problem in using.

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