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How effective is ultrasound-based screening for trisomy 18 without the addition of biochemistry at the time of late first trimester?

Abstract

Trisomy 18 (T18) remains the second most common aneuploidy. It is associated with multiple congenital anomalies and causes intrauterine fetal demise in the most severe cases.
Objectives: To examine the screening performance of ultrasound-based protocols for detecting T18, we aimed to determine the most common signs and their prevalence in fetuses with T18 to develop logistic regression model.
Methods: This was a prospective study based on singleton pregnancies examined at gestation 11+0 to 13+6. The referrals constituted 6210 patients. Scan protocol enclosed a systematic review of the entire early fetal anatomy, including fetal cardiac evaluation and sonographic signs of aneuploidy.
Results: Our study population comprised 5650 pregnancies: 5613 cases with a normal karyotype and 37 cases with T18. The mean nuchal translucency (NT) thickness in the subgroup of euploidy was 1.7 and in the subgroup of T18 it was 5.4. No statistically significant differences were found in terms of maternal age. One case of T18 (2.7%) demonstrated no markers of aneuploidy as opposed to 5111 cases of euploidy (91.1%). Extracardiac malformations were identified in 13 cases of T18 (35.1%) and in 48 cases of euploidy (0.8%). Congenital heart defects were observed in 26 cases of T18 (70.3%) and in 27 cases of euploidy (0.5%).
Conclusions: Our results showed good screening performance of ultrasound-based risk calculation models. When the first trimester pattern of T18 is considered, an increased NT, tricuspid regurgitation, single umbilical artery, omphalocele and right dominant heart should be specifically searched for.

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Keywords: Chromosomal aberrations; first trimester; trisomy 18 (T18).

DOI 10.1515/jpm-2014-0384
Received December 20, 2014. Accepted February 25, 2015.

Introduction

Trisomy 18 (T18) also called as Edwards syndrome (ES) remains the second most common aneuploidy. It is associated with multiple congenital anomalies, causes intrauterine fetal demise (IUFD) in the most severe cases and is responsible for early neonatal deaths in affected liveborns. The total prevalence of T18 is 5.4 per 10,000 births, which increases with maternal age (MA) reaching 61.93 per 10,000 births in mothers over 40 years [1]. Prenatal diagnostic features of ES in mid-pregnancy have been well described in the literature and have proved high detection rate (DR) for this anomaly due to the syndromal pattern indicative for T18 [2–6]. Evaluation of early anatomy at the time of gestation 11+0 to 13+6 weeks is feasible as well as many of the affected fetuses present abnormal sonographic findings at this time, e.g., omphalocele (OMPH), abnormal posturing of the hands, megacystis, congenital heart defects (CHDs) [7, 8]. With the introduction of first trimester screening based on MA, fetal nuchal translucency (NT), free β-hCG and pregnancy-associated plasma protein-A (PAPP-A) in a routine antenatal care, the diagnosis of this condition has been possible to be made at an earlier gestational age [9–12]. However, with the use of this so-called combined screening test (CST), DRs vary according to the literature [10, 12–14]. It has been estimated that the CST protocol for trisomy 21 (T21) identifies 82% fetuses with T18 at an false-positive rate (FPR) of 3% [10]. However, the use of a specific risk algorithm for T18 has a variable DR between 79% and 93% at an FPR ranging from 0.2% to 0.5% according to different studies [10, 12–14]. Addition of secondary ultrasound markers (CST+) like nasal bone (NB), tricuspid regurgitation (TR) and ductus venosus (DV) flow to

the first trimester combined screening that have been proved to enhance the DRs of T21 [15–17] provided divergent results for identifying fetuses with T18. Sensitivity of screening performance based on this approach varied between 89% [15] and 100% [16] with an FPR of 3.4 [15] to 4.8 [16]. Despite improvements in early diagnosis of T18, there is a paucity of studies describing the effectiveness of screening solely based on ultrasound that would include all secondary markers of aneuploidy in addition to NT. The main objective of the study was to examine the screening performance of protocols dedicated for T21 and T18 used in detecting T18 by operating just ultrasound-based parameters established on primary (NT) and also secondary (NT+) markers of aneuploidy (NT, NB, TR, DV) enhanced with early anomaly and early echocardiography findings and to assess whether the performance of the method depends on MA ranges. Furthermore, we aimed to determine the most common signs and their prevalence in fetuses with ES to develop logistic regression model.

Methods

This was a prospective study based on singleton pregnancies examined at gestation 11+0 to 13+6 weeks at our institution. The referrals constituted of low-risk patients (4895) and a weighty set of high-risk cases (1315). High risk subjects included patients with MA above 35 years (783) and cases presenting suspicious ultrasound findings on the initial scan performed by non-qualified for first trimester screening obstetricians (532). Karyotyping results and postnatal evaluation findings were covered in the database as soon as they were accessible. Patients, who were examined between January 2009 and June 2012, were included. The sonography reports together with digital data were reviewed taking into account the following inclusion criteria: singleton pregnancy, crown-rump length (CRL) measurement of 45–84 mm, known pregnancy outcome. The patients' body mass index (BMI) was computed in kilogram per square meter on the day of the late first trimester ultrasound scan. Fetal karyotyping was evaluated from amniotic fluid samples (653 cases). The rest of the subjects were measured to be euploid, based on normal postnatal evaluation. Aneuploidies, other than T18, were excluded from the exploration. Three examiners qualified for the complete set of sonographic markers by the Fetal Medicine Foundation (FMF) were engaged in this study. One examiner had 1 year (AM) and two others had 8 years of experience in the first trimester screening (MW and AN). All scans were performed utilising the Voluson E6 ultrasound scanner (GE Healthcare, Zipf, Austria) from transabdominal approach. In 5.4% of ultrasound scans transvaginal probe was applied for better definition of fetal anatomy. Scan protocol enclosed a systematic review of the entire early fetal anatomy, including fetal cardiac evaluation based on the following parameters: visceral situs, four-chamber view (4CV), outflow tracts, three-vessel and trachea view in B-mode and color mapping. The sonographic signs of chromosomal aberrations (NT, NB, TR, DV) were checked following the FMF recommendations. DV was evaluated by a qualitative method. The sonographic findings among euploidy and T18 were investigated. The history and ultrasound observations and measurements were utilized for T18 risk calculations by using FMF 2.3.2 software (Astraia Gmbh, Munich, Germany). In all cases to detect ES cases, four ultrasound-based risk calculation methods were applied: "adjusted risk for trisomy 18 by NT","adjusted risk for trisomy 18 by NT+", "adjusted risk for T21 by NT" and "adjusted risk for T21 by NT+". In the second and fourth methods all secondary ultrasound markers (NB, TR, DV) were employed together with NT. Adjusted risk for T18 or 21 above 1/100 at the time of scans was defined as a high-risk, independently on the method. All high-risk subjects and cases with detected structural malformations but presenting low-risk figures had genetic counselling and underwent sonography between 18 and 19 weeks of gestation according to AIUM second trimester and fetal echocardiography recommendations [18, 19]. The outcome data were collected from medical records and also included karyotyping, 18–21 and 28–32 weeks of gestation ultrasound, autopsy and neonatal findings. The local Ethics Committee approved the study protocol and all subjects gave written consent.

Statistical analysis

In statistical data assessment, the Kolmogorov-Smirnov test was applied for continuous variable distribution. The χ^2 test was used to demonstrate the differences. Groups of independent variables were compared using the Student's *t*-test. Non-parametric tests were also utilised. SPSS Statistics v.17 (IBM Co., New York, NY, USA) software was applied in this study. The figures of $P<0.05$ were measured as significant. Radar logarithmic charts were used to visualise screening uptake. From the most frequent features apparent in our group of fetuses with T18, logistic regression model was developed, which was used to calculate odds ratios (ORs) for the commonest ultrasound findings.

Results

Study population

Screening ultrasound was carried out in 6210 singleton pregnancies. However, 560 (9.0%) cases were excluded from further analysis because in 380 (6.1%) cases it was impossible to establish the fetal karyotype due to losing them from the follow-up, 43 (0.7%) cases resulted in miscarriages not related to invasive testing and 23 (0.4%) cases with IUFD without subsequent karyotyping; in 114 (1.8%) cases there was a chromosomal abnormality other than T18. Therefore, our study population comprised 5650 pregnancies: 5613 cases with a normal karyotype or delivery of a normal baby (euploid group) and 37 cases with T18. The characteristic of the study population is summarized in Figure 1. The median maternal BMI was 22.4 kg/m^2 (range 17.6–35.2). All women participating in this study were Caucasians.

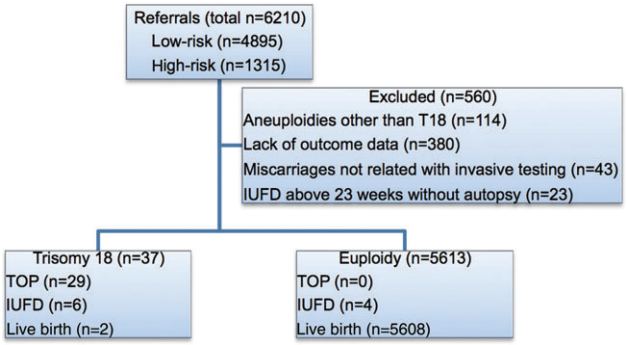

Figure 1: Study population diagram.
T18=trisomy 18, IUFD=intrauterine fetal demise, TOP=termination of pregnancy.

The mean NT thickness in the subgroup of euploidy was 1.7 mm (range 0.1–4.9 mm) and in the subgroup of T18 it was 5.4 mm (range 1.4–11.7 mm) (P<0.005). The mean MA in the euploid group was 30.5 years (range 25–42 years) compared to 33.3 years (range 18–46 years) in T18 (P=0.162). The mean CRL at the time of examination was 63.3 mm in euploid vs. 61.1 mm in T18 group. No statistically significant differences were found between euploidy and T18 groups in terms of MA, CRL and fetal heart rate (FHR) (Table 1).

The NT thickness above the 95th percentile was observed in 228 euploid fetuses (4.06%) and in 31 fetuses (83.7%) affected by T18 (Figure 2). By using χ^2 Pearson test statistical differences were found in the presence of TR (P=0.000) and reverse a-wave in DV (revDV) flow (P=0.000) between the groups of euploidy and T18.

Only 1 case of T18 (2.7%) demonstrated no markers of aneuploidy as opposed to 5110 cases of euploidy (91.1%).

Table 1: Comparison of fetuses with euploidy and trisomy 18 according to four parameters.

Karyotype	CRL (mm)	NT (mm)	FHR (bpm)	MA (years)
Euploidy				
n	5613.0	5613.0	5613.0	5613.0
Mean	63.3	1.7	160.3	30.5
Median	62.8	1.6	160.0	30.0
SD	9.1	0.5	7.3	4.2
Trisomy 18				
n	37.0	37.0	37.0	37.0
Mean	61.1	5.4	158.4	33.3
Median	59	5.1	158.0	34.0
SD	9.7	2.5	9.5	6.8
Statistical significance	0.162	0.000	0.162	0.008

CRL=crown-rump length, NT=nuchal translucency, FHR=fetal heart rate, MA=maternal age, SD=standard deviation.

Isolated markers were identified in two cases of T18 (5.4%) including one case with increased NT above the 95th percentile and one case with TR. Isolated markers were identified in 419 cases of euploidy (7.8%) including subjects with: NT above the 95th percentile (181 cases); delayed nasal ossification (63); TR (71) reversed a-wave in DV flow (94); single umbilical artery (SUA) (6); and absent DV in four cases. SUA was noted in six cases of euploidy (0.1%) and in 20 subjects of T18 (54.0%); this difference was statistically significant (P<005). The commonest core coincidences of aneuploidy ultrasound markers in T18 were NT above the 95th percentile with TR, noted in 16 cases (43.2%) and NT above the 95th percentile with delayed nasal ossification, found in 11 cases (40.7%). These core combinations were observed in euploidy only in 18 (0.3%) and 17 (0.3%) cases of euploidy, respectively. The details showing configuration of ultrasound markers of aneuploidy in the group of euploidy and T18 are summarised in Table 2.

Extracardiac malformations (ECMs) were identified in 13 cases of T18 (35.1%) and in 48 cases of euploidy (0.8%). This difference was statistically significant (P<0.005). ECMs were found isolated among all euploidy cases. This was also the case in T18 except for one case of exomphalos containing bowels combined with congenital diaphragmatic hernia. Details are depicted in Table 3.

Regarding CHDs we observed these defects in 26 cases of T18 (70.3%) and in 27 cases of euploidy (0.5%). The largest fractions of CHDs in T18 were: ventricular septal defects (VSDs)—32.4%, cases with double outlet right ventricle (DORV)—16.2% and hypoplastic left heart syndrome (HLHS)—8.1%. Details are depicted in Table 4.

The most prevalent ultrasound findings in T18 cases were computed by logistic regression to demonstrate their ORs. In this analysis the following parameters were included: NT>95th percentile; TR; SUA; right dominant heart (RDH) as a sign of HLHS and DORV; and the presence of OMPH (Table 5). We excluded VSDs from the regression model due to the high risk of false-positive results. The interventricular septum requires better resolution and the late first trimester scan is still not conclusive for VSDs [20].

Screening performance

Actual screening efficacy for detecting T18 by the risk cut-off above 1/100 of NT-only protocols ("adjusted risk for T18 by NT" and "adjusted risk for T21 by NT"), NT enhanced with secondary ultrasound markers protocols ("adjusted risk for T18 by NT+" and "adjusted risk for T21 by NT+") and simple methods like MA, NT above the 95th

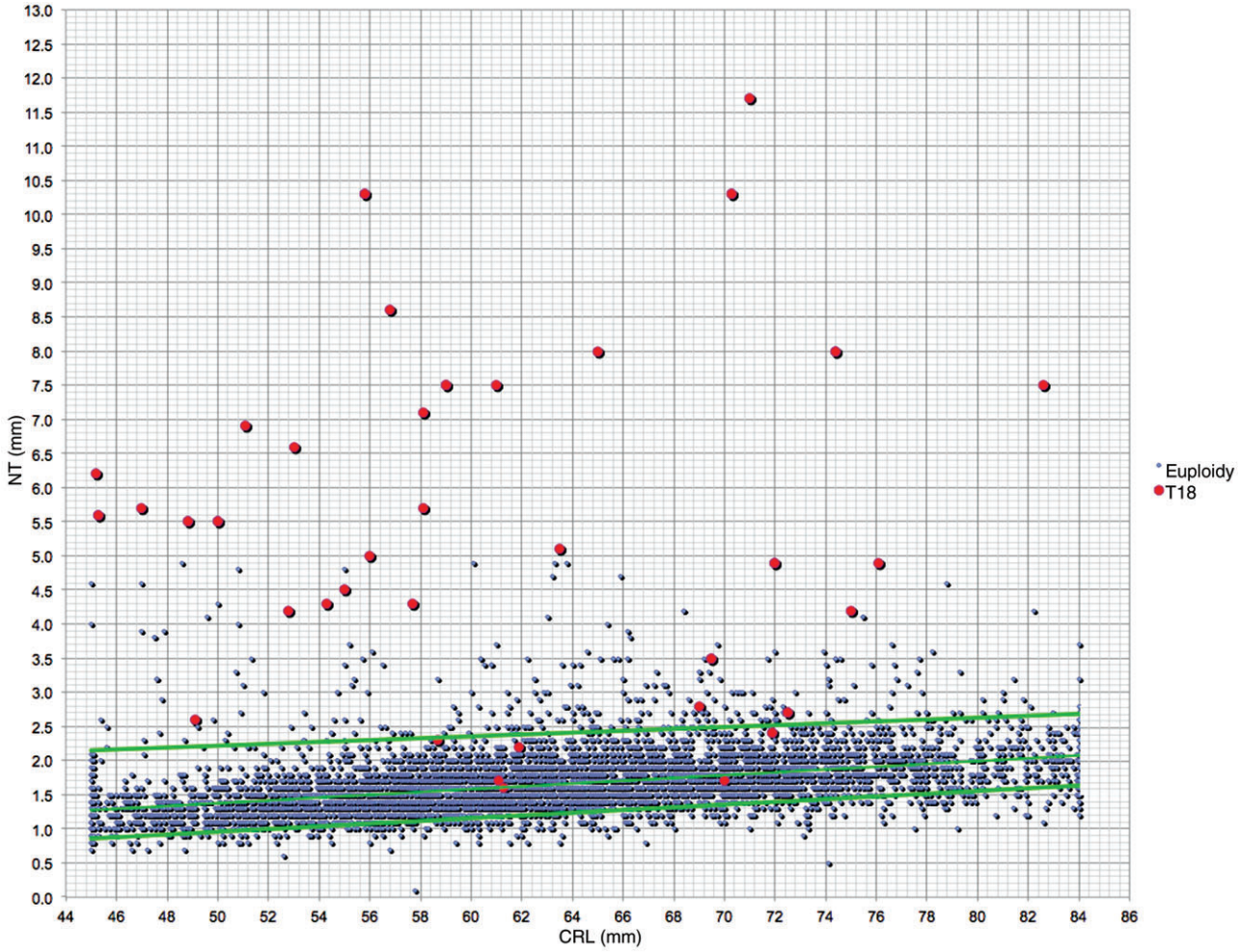

Figure 2: Distribution of fetal nuchal translucency (NT) thickness according to crown-rump length (CRL) in euploid fetuses (blue dots) and in cases with trisomy 18 (red dots).

percentile, absent NB, presence of TR, SUA and the evidence of revDV are summarized in Table 6.

Radar-type logarithmic charts were used to visualize the number of screen-negative T18 cases depicted as dots seen out of the blue area of high risk in four adjusted methods (Figure 3).

The receiver operating characteristics (ROC) of NT-only and NT-enhanced protocols are depicted in Figure 4.

On the basis of ROC curves, for a given arbitrary FPR of 3%, the DR for T18 by using "adjusted risk for T21 by NT" protocol was 78% (95% confidence interval, CI, 71.6–82.4) compared to 92% (95% CI, 85.1–98.0) for "adjusted risk for T21 by NT+". For a given arbitrary FPR of 3%, the DR for T18 by using "adjusted risk for T18 by NT" protocol was 84% (95% CI, 77.4–100.6) compared to 95% (95 CI, 88.0–102.0.0) for "adjusted risk for T18 by NT+". These results are shown in Table 7 and compared with DRs at the given FPR of 5%.

T18 detection rate in relation to maternal age

We noted 13 cases of T18 in MA range between 26 and 30 years; six cases in MA range between 31 and 35 years; 14 cases in MA range between 36 and 40 years; and four cases above 40 years. There was a tendency of increase in DR and FPR with the advance of MA in two analysed risk calculation methods. However, models: "adjusted risk for T21 by NT+", "adjusted risk for T18 by NT" and "adjusted risk for T18 by NT+" demonstrated the lowest DR in the MA range between 31 and 35 years. The model "adjusted risk for T21 by NT" showed the lowest DR in

Table 2: Configuration and prevalence of isolated and combined markers of aneuploidy in euploid and trisomy 18 fetuses.

Karyotype	n	%
Euploidy		
No markers	5110	91.1
NT	181	3.2
NB(–)	63	1.1
TR	71	1.3
revDV	94	1.7
noDV	4	0.1
SUA	6	0.1
NT+NB(–)	7	0.1
NT+TR	10	0.2
NT+revDV	14	0.2
NT+NB(–)+TR	5	0.1
NT+NB(–)+revDV	3	0.1
NT+NB(–)+TR+revDV	2	0.0
NB(–)+revDV	7	0.1
NT+0DV	2	0.1
NB(–)+TR	6	0.2
TR+revDV	1	0.1
NT+TR+revDV	1	0.1
Trisomy 18		
No markers	1	2.7
NT	1	2.7
TR	1	2.7
NT+NB	4	10.8
NT+TR	7	18.9
NT+revDV	6	16.2
NT+NB+TR	4	10.8
NT+NB+revDV	3	8.1
NB+revDV	1	2.7
NB+TR	1	2.7
NT+TR+revDV	5	13.5
TR+SUA	2	5.4
DV+SUA	1	2.7

NT=nuchal translucency above the 95th percentile, TR=tricuspid regurgitation, NB(–)=negative nasal bone, revDV=reversed a-wave in ductus venosus flow, noDV=absent ductus venosus, SUA=single umbilical artery.

the age group 36–40 years. The results of analysis of DR and FPR depending on MA ranges are presented in Figure 5.

Discussion

This study describes our experience with the detection of T18 at the time of first trimester scan. Our results showed good screening performance of ultrasound-based risk calculation models at a cut-off of 1/100. The specific risk algorithm for T18 based on the complete set of ultrasound markers of aneuploidy ("adjusted risk for T18 by NT+") was the most sensitive one and identified about 95% of affected fetuses at an FPR of 1.2%. The risk algorithm for

Table 3: Extracardiac structural abnormalities summarized in terms of chromosomal status.

Karyotype	n	%
Euploidy		
No ECM	5565	99.0
Hydrops	5	0.1
Brain anomalies	12	0.2
Abdominal anomalies	14	0.2
Urinary tract anomalies	15	0.3
Limb anomalies	15	0.3
Facial and neck anomalies	9	0.2
Thoracic anomalies	3	0.0
Trisomy 18		
No ECM	24	64.9
Hydrops	2	5.4
Abdominal anomalies	8	21.6
Urinary tract anomalies	1	2.7
Limb anomalies	2	5.4
Thoracic anomalies	1	2.7

Table 4: Cardiac anomalies summarized in the group of euploidy and trisomy 18.

Karyotype	n	%
Euploidy		
No CHD	5586	99.5
Septal defects	2	0.0
Conotruncal anomalies	10	0.2
Left heart defects	7	0.1
Right heart defects	5	0.1
Heterotaxy	1	0.0
Aortic arch defects	1	0.0
Trisomy 18		
No CHD	11	29.7
Septal defects	12	32.4
Conotruncal anomalies	7	18.9
Left heart defects	3	8.1
Aortic arch defects	1	2.7
Cardiomegaly	2	5.4
Right heart defects	1	2.7

CHD=congenital heart defect.

Table 5: Odds ratios (ORs) for the commonest features determined in trisomy 18 fetuses based on logistic regression model.

	P	OR	95% CI for OR	
			Lower limit	Upper limit
NT>95th per	0.000	6.0	3.5	10.2
TR	0.000	63.6	13.4	301.4
SUA	0.001	23.9	3.4	165.5
RDH	0.000	134.6	11.7	1548.4
OMPH	0.000	50.7	7.6	335.9

TR=tricuspid regurgitation, SUA=single umbilical artery, RDH=right dominant heart, OMPH=omphalocele.

Table 6: Screening performance summary of the methods used in this study (in parentheses the estimates of lower to upper 95% CIs are shown).

Screening method	Adjusted risk for T18 by NT	Adjusted risk for T18 by NT+	Adjusted risk for T21 by NT	Adjusted risk for T21 by NT+	MA>35 years	NT>95th percentile	NB	TR	revDV
Sensitivity	75.7% (59.8–86.6)	94.6% (82.3–98.5)	91.9% (78.7–97.2)	78.4% (62.8–88.6)	45.9% (31.0–61.6)	83.8% (68.9–92.3)	35.1% (21.8–51.2)	54.0% (38.4–68.9)	48.6% (32.9–64.4)
Specificity	99.2% (98.9–99.4)	98.9% (98.6–99.1)	96.7% (96.2–97.2)	97.2% (96.2–97.2)	83.6% (82.6–84.5)	94.1% (93.4–94.7)	98.2% (97.9–98.5)	99.4% (99.2–99.6)	97.9% (97.5–8.3)
PPV	37.3% (27.3–48.6)	36.1% (27.2–46.0)	15.7% (11.4–21.1)	15.5% (11.0–21.4)	1.8% (1.1–2.9)	8.54% (6.1–11.9)	11.6% (6.9–18.8)	39.2% (27.0–52.9)	12.8% (8.1–19.5)
NPV	99.8% (99.7–99.9)	99.9% (99.9–99.9)	99.9% (99.8–99.9)	99.8% (99.7–99.9)	99.6% (99.3–99.7)	99.9% (99.7–99.9)	99.6% (97.4–99.7)	99.7% (99.5–99.8)	99.7% (99.5–99.8)
Diagnostic accuracy	99.0 (98.7–99.2)	98.8% (98.5–99.1)	96.7% (96.2–97.1)	97.0% (96.6–97.5)	83.4% (82.4–84.3)	94.0% (93.4–94.6)	97.8% (97.4–98.2)	99.1% (98.9–99.4)	97.6% (97.2–97.9)

T18=trisomy 18, T21=trisomy 21, PPV=positive predictive value, NPV=negative predictive value, NB=negative nasal bone, TR=tricuspid regurgitation, revDV=reversed a-wave in ductus venosus flow.

T21 (adjusted risk for T21 by NT+) showed slightly worse performance of 92% in detection of this anomaly. We found that the MA had a strong positive influence on DR and FPR independently on the risk calculation models. Surprisingly, a drop of DR to 85.7% was observed in the MA range of 31–35 years in the most efficient model ("adjusted risk for T18 by NT+"), but in this range only six cases were included. The advantage of this study is the fact that we evaluated screening methods for T18 based only on ultrasound findings as to best of our knowledge; there is a lack of publications concentrating on this subject at the time of first trimester scan. The possible disadvantage is the fact that the ultrasound-based method is only effective when all sonographic markers are analyzed and the examiner is certified for the complete package of these markers. General screening practices do not cover the application of secondary markers, especially the cardiovascular ones. According to the actual data acquired from FMF website general screening ability varies among countries. Out of active first trimester screening healthcare providers, who are regularly audited for NT, 67% were certified for TR in Germany, 69% in Poland, 48% in the UK and 17% in the USA. Certification for DV in the same group shows following figures: Germany (71%), Poland (78%), the UK (46%) and the USA (13%). The results of our study demonstrate that it is worth investing in training of sonographers and physicians in secondary first trimester ultrasound markers to achieve better results in screening. Another disadvantage of our analysis may be our policy of focusing on T18 cases by excluding other aneuploidies. This approach could reduce the number of subjects of various chromosomal aberrations demonstrating overlapping features, but without this extraction the commonest markers of T18 would not be so clearly highlighted. The mixed risk referral population used in our study reflects local OB/GYN healthcare practice, which covers first trimester basic sonogram focused on viability and fetal growth at every antenatal care provider in Poland. Due to this fact tertiary screening centres note greater number of high-risk patients, i.e., with suspicion of thickened NT from non-qualified for screening referring physicians. In our opinion it only bears the risk of higher FPR and would not influence on DR. The population included in our study comprised women of Caucasian origin only; therefore, the results and conclusions cannot be drawn for patients of other racial background.

Due to the handful data available in the literature regarding efficacy of the ultrasound-only based screening algorithms in the detection of T18, we must compare our results with the observations drawn from CST and CST+ studies, although in our models none of the biochemical assays was employed [10, 15, 16]. So far, specific

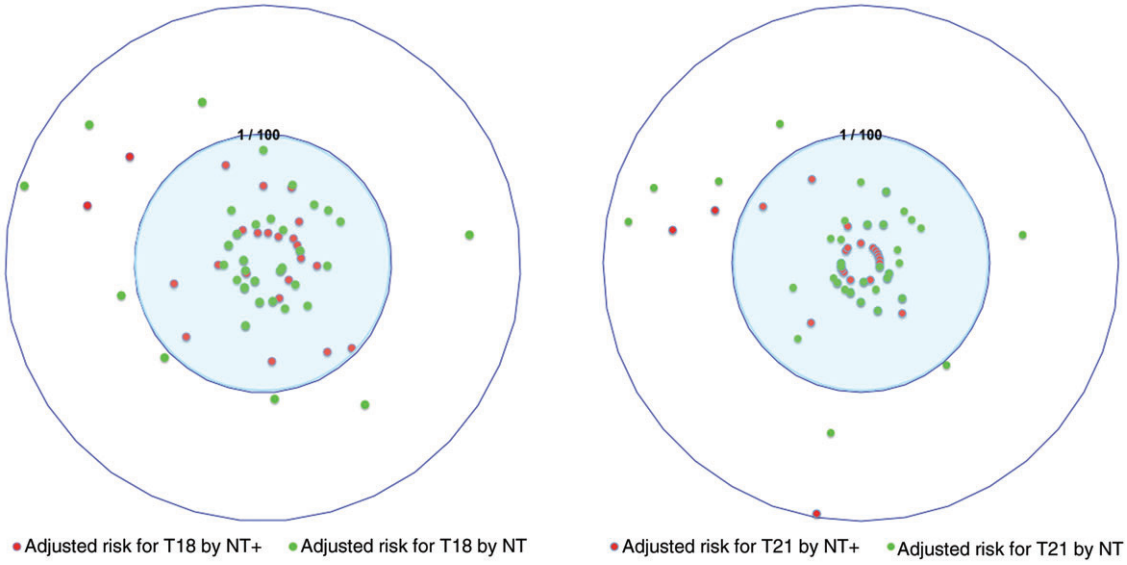

Figure 3: Radar logarithmic charts showing the uptake of four ultrasound-based screening methods to detect trisomy 18 (T18). "Adjusted risk for T18 by NT" presented with green dots (left) and "adjusted risk for T18 by NT+" presented with red dots (right). "Adjusted risk for trisomy 21 (T21) by NT" presented with green dots and "adjusted risk for T21 by NT+" presented with red dots.

risk algorithms for T18 based on MA, fetal NT, free β-HCG and PAPP-A demonstrated different DRs between 79% and 93% at an FPR 0.2–0.5 [10, 12–14]. In first trimester screening for T21 the performance of the CST has been improved by the inclusion of additional ultrasound markers such as absent NB, reversed end-diastolic flow in DV and TR. However, integrating the above-mentioned markers in detection of T18, which are also commonly found in affected fetuses, provided different results. Ghaffari et al. identified 100% of fetuses with T18 at an

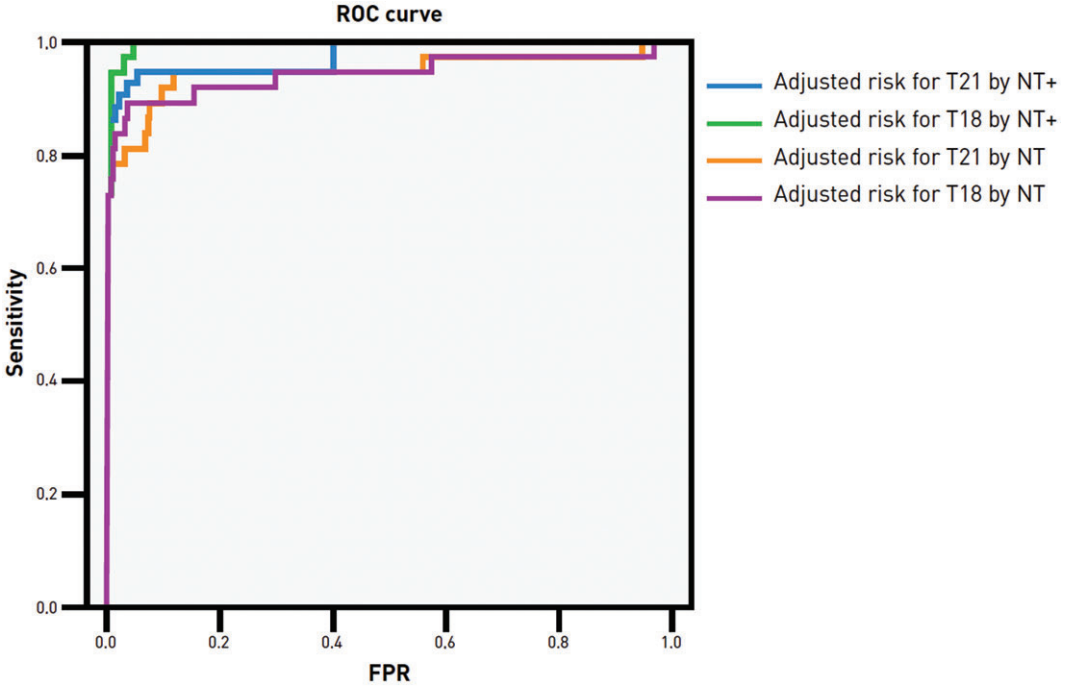

Figure 4: Effectiveness of screening for trisomy 18 by using receiver operating characteristic curves. "Adjusted risk for T21 by NT+" method – blue line with the area under the curve (AUC)=0.954. "Adjusted risk for T21 by NT " method – orange line with AUC=0.984. "Adjusted risk for T18 by NT+" method – violet line with AUC=0.984.

Table 7: Detection rates (DR) and 95% CIs at fixed false-positive rates (FPR) of 3% and 5% in four adjusted risk screening methods.

Screening test	AUC	DR% (95% CI) at 3% FPR	DR% (95% CI) at 5% FPR
DR of "adjusted risk for T21 by NT+"	0.983	92% (85.1–98.0)	95% (88.0–102.0)
DR of "adjusted risk for T18 by NT+"	0.995	95% (88.0–102.0)	100% (92.8–107.2)
DR of "adjusted risk for T21 by NT"	0.945	78% (71.6–82.4)	81% (74.5–87.5)
DR of "adjusted risk for T18 by NT"	0.942	84% (77.4–100.6)	89% (82.2–97.8)

AUC=area under the curve.

FPR of 3.44; however, only two cases were included in the analysis [15]. In another study DR of T18 based on combined screening and secondary ultrasound markers was estimated at 89% at FPR of 4.8 [16]. In our analysis we did not incorporate biochemical testing in the analysis. It was previously found that serum levels of PAPP-A and free β-HCG are affected by maternal characteristics, including racial origin, weight, smoking and method of conception as well as the machine and reagents used for the analysis. Furthermore, to provide the most reliable results of the screening, biochemical testing and ultrasound scanning should be carried out at best in two separate visits, with the first one done at 9–10 weeks and the second at 12 weeks, which in our conditions could not be offered to the patients [21–23]. According to our study increased NT above the 95th percentile was present in 83.7% cases of T18 and in our regression model this parameter shows the OR for T18 of 6. Our observations are in line with the results of previous studies [7, 8, 24, 25] and the most striking first trimester sonographic feature of this condition was the increased NT thickness, which was usually prominent and frequently associated with subcutaneous edema. All but one fetus displayed one or more abnormal sonographic marker, most commonly in combinations (94.6%) such as NT>95th percentile plus TR (43.2%) and NT>95th percentile and delayed nasal ossification (40.7%). In a regression model TR raised the OR for T18 of 63.6. Of the structural anomalies detected in our cases the most common were cardiac defects (70.3%) especially VSDs and RDH. This compares well with other reports where the frequency of cardiac defects was estimated at approximately 83%–84% [24, 26] with VSDs found in the majority of cases. Despite that VSD was the most frequent anomaly observed in our series as we excluded this defect from the regression model due to the high risk of false-positive results. The interventricular septum requires better resolution and the late first trimester scan is still not conclusive for VSDs [20]. However, we suggest taking particular care for RDH due to the fact, that DORV, coarctation of aorta, mitral atresia and HLHS constitute frequent anomalies in T18 [27–29]. Logistic regression model used in our study showed OR of 134.6 for RDH. Simple color mapping at the level of 4CV is useful in our opinion to search for RDH, which can be done just after the sampling of tricuspid valve in pulsed-wave Doppler (Figure 6).

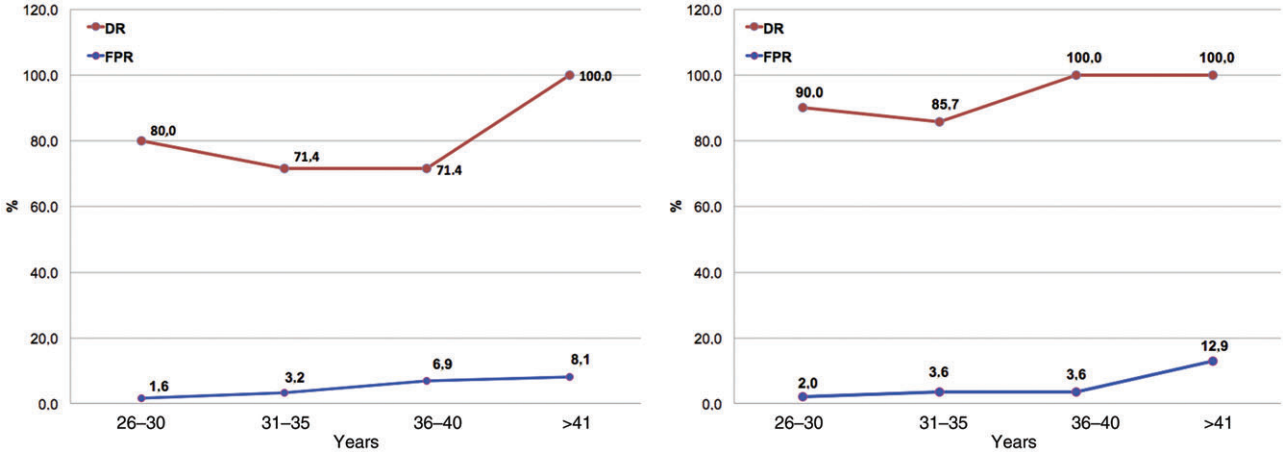

Figure 5: Detection rates (DRs) and false-positive rates (FPRs) of the "adjusted risk for T21 by NT" depending on maternal age (MA) ranges (left). DRs and FPRs of the remaining methods: "adjusted risk for T21 by NT+", "adjusted risk for T18 by NT" and "adjusted risk for T18 by NT+" depending on MA ranges demonstrate the same values (right).

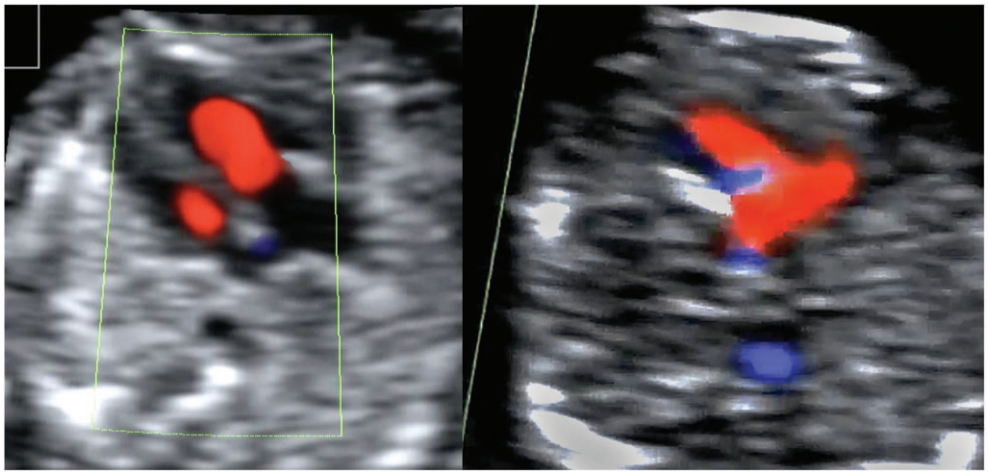

Figure 6: Early fetal echocardiograms in color Doppler mapping depicting trisomy 18 cases, which were included in the group of right dominant heart. On the left the stronger inflow is seen to the ventricle, which is closer to the anterior thoracic wall in case of double outlet right ventricle. On the right, only one inflow is observed together with a hyperechoic area seen to the left of this inflow in case of hypoplastic left ventricle.

Our observations are similar to those published by Cheng et al., who proved that fetal NT measurement together with fetal echocardiography between 16 and 18 weeks is a sensitive method for detecting T18 [28]. Regarding the heart examination, bradycardia is a finding that has been associated with T18 [8]. It was suggested that it may represent a preterminal decompensation of a circulatory system, in part explaining an increased NT, being representative for the cases at greater risk for an early intrauterine demise [7]. Although in our study we did not observe any significant differences in mean FHR between euploidy and T18, FHR values below the 5th percentile were found in a small proportion of cases similar to that of 18.7% reported by Liao et al. [30].

As it was raised during earlier observations, we confirmed that the check-up for SUA is important when T18 is suspected [31]. It was identified in 54% of ES cases and showed an OR of 23.9 in logistic regression. Rembouskos showed higher prevalence of SUA in T18 at a level of 77.8% [31]. On the contrary, Sepulveda et al. [7] did not record this anomaly within the group of fetuses with T18, but these researchers admitted that this anomaly was not specifically looked for. Yeo et al. has examined 38 fetuses affected by T18 in the second trimester and demonstrated that the number of abnormalities per fetus was eight on average, and four at a minimum [26]. In the review of extracardiac anatomy our cases of T18 demonstrated mainly isolated defects, which were observed in 35.1% of cases, with OMPH as the most common (21.6%; OR—50.7). Our results are indeed consistent with those from Sepulveda et al. [7] in which the most prevalent structural anomaly associated with Edwards syndrome (ES) was OMPH with a rate of 21%. Despite similar characteristics of population screened, which were derived from a referral centre, the frequency of exomphalos in our series is lowered to 26% as reported by Sherod et al. [8]. In counseling of the patients it is important to note that differential diagnosis should include physiological midgut herniation, which is expected to regress before 11 weeks [32]. A thorough examination of the fetus should be performed as exomphalos associated with a major structural defect or an increased NT in the first trimester implies a high risk of aneuploidy of 78.9% and 72.2%, respectively, with T18 comprising 72% of cases [33]. Parents can be reassured that the fetuses with isolated exomphalos and normal NT are likely to be euploid. Remarkably, only one fetus was noted to have a megacystis in the first trimester, although this anomaly is almost always invariably present in fetuses with ES. Similarly, abnormal posturing of the hands, one of the most common morphological second trimester findings for T18, was noted in only two cases (5.4%). One explanation may be that this subtle anomaly may be more challenging to visualise in the first trimester. However, introduction of 3D technique enables better imaging of limb defects before 15 weeks of gestation [34] demonstrating that fetuses with T18 have narrower and shorter hands compared to the euploid ones [35]. A possible explanation of misdiagnosis of clenched hands may be the fact that in some cases they may appear later in gestation. Quintero et al. have reported a fetus, evaluated under fetoscopy, which did not demonstrate clenched hands before the end of the 13 weeks of gestation [36].

According to the literature there is a scarce data regarding the possible influence of MA on performance of the first trimester screening method. We regard this topic as very important as the time of childbearing has changed over years. Artificial reproductive techniques give further chances for pregnancy in later reproductive age. Interestingly, we found that the sensitivity of the screening decreased when MA was in the range of 30–35 years. Although DRs of T18 increased in age ranges 36–40 and above, FPR increased as well. This would imply the risk of overestimation and unnecessary invasive procedures in older women whose pregnancies are most often due to IVF procedures. However, for a proper counseling, these findings should be interpreted with caution and future studies investigating more pregnancies are required to produce more precise information on the dependence of screening performance of the first trimester scan on MA.

In summary, our study showed a good screening performance of ultrasound-based methods including all markers of aneuploidy and anomaly scan. We confirm that the DR is dependent on the MA; however, further studies comprising bigger population should be performed. When the first trimester pattern of T18 is considered, an increased NT, TR, SUA, OMPH and RDH should be specifically searched for.

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The authors stated that there are no conflicts of interest regarding the publication of this article.