Clinical laboratory experience with noninvasive prenatal testing in twin gestations

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Abstract

Introduction: The verifi® test has been available for twin gestations through the Illumina CLIA lab since October 2013. Published studies of NIPT in twin gestations are limited to small numbers. This study summarizes clinical lab metrics of NIPT in >1000 twin gestations.

Methods: Clinical lab data from twin pregnancies were reviewed. Pregnancy outcome for cases with Aneuploidy Detected (AD) or Aneuploidy Suspected (AS) result was collected following an established protocol.

Results: During the study period, 1,456 samples from twin gestations were received from US and non-US sites for testing. Zygosity was not reported for most. Results were reported in 1440 cases (98.9%) with average turnaround time of 3.1 business days. Of 16 (1.1%) total cancellations, one (0.07%) was due to technical reasons. Median gestational age was 12 wks (range 10-36 wks) and mean maternal age was 35.9 yrs. Aneuploidy was detected in 27 (1.88%); 24 were T21 (1.67%) and 2 were T13 (0.14%). One case was AD for full or partial monosomy 18. Four cases (0.28%) were AS [2 T21, 1 T18, and one double aneuploidy AS T18 and AD T21]. Sex chromosome analysis, reported as presence or absence of Y chromosome, was requested in 1318 cases; Y was present in 930 (70.6%) and absent in 388 (29.4%). Outcome was available in 46.6% (14) of cases with AD or AS results. Outcome information was unavailable mainly from non-US samples. Eleven AD cases were concordant. There were 3 (0.21%) putative false positives (1 AS T21, 1 AD T21, 1 AD T13) and no known false negatives.

Conclusions: This study demonstrates that the verifi® test algorithm for twin gestations shows excellent test metrics in a clinical lab setting. The patterns seen in this study are similar to our previously reported clinical lab experience for singleton gestations (see Table), providing further support for the utility of NIPT in twin pregnancies. Future studies analyzing pregnancy outcomes in twin gestations are needed to better define clinical test performance of NIPT in this population.

	Singletons	Twins
TAT	3.9	3.1
Cancellation %	0.1%	0.07%
Average MA	35.4	35.9
Average GA	14.2	13.94
Aneuploidy Detected/ Suspected	3.17%	1.88%

Background

- The sensitivity of standard maternal serum aneuploidy screening is reduced in twin gestations.
- The risk of miscarriage associated with invasive diagnostic procedures is higher in twin gestations than in singleton pregnancies.
- Thus, prenatal testing options for twin pregnancies are limited.
- The verifi® test through the Illumina CLIA lab has been available for singleton gestations since March 2012 and expanded to twin gestations in October 2013.
- Published studies of noninvasive prenatal testing (NIPT) in twin gestations are limited to small numbers.

Study Objective

- To summarize the experience of a single laboratory performing aneuploidy screening for trisomy 21, 18, and 13 via genome-wide massively parallel sequencing of cell free DNA in maternal plasma in samples received from twin gestations.
- To provide clinically relevant metrics for the verifi® Prenatal Test in twin gestations and compare these metrics to singletons[1].

Methods

- Test requisition forms where twin pregnancy was indicated by the ordering provider were reviewed.
- Demographic information including maternal age and gestational age at draw, as well as clinically relevant metrics such as
  turn-around time, from 1456 twin pregnancies were reviewed and compared to singleton pregnancies[1].
- Clinical outcome for cases with Aneuploidy Detected (AD) or Aneuploidy Suspected (AS) result was collected following an established laboratory protocol.

Results

Table 1. Comparison of Current Study Lab Metrics to Clinical Lab Metrics with Singletons

	Current Study	Clinical Experience with singletons[1]
Total cases	1,456	34,860
Average Turn-around Time	3.1 business days	3.9 business days
Total Cancellations	16 (1.1%)	554 (1.6%)
Technical[a]	1 (0.07%)	39 (0.1%)
Administrative[b]	15 (1.0%)	515 (1.5%)

[a] Technical cancellations include samples that did not meet quality control standards

[b] Administrative cancellations include samples that did not begin the testing process

Table 2. Comparison of Current Study Demographics to Clinical Experience with Singletons

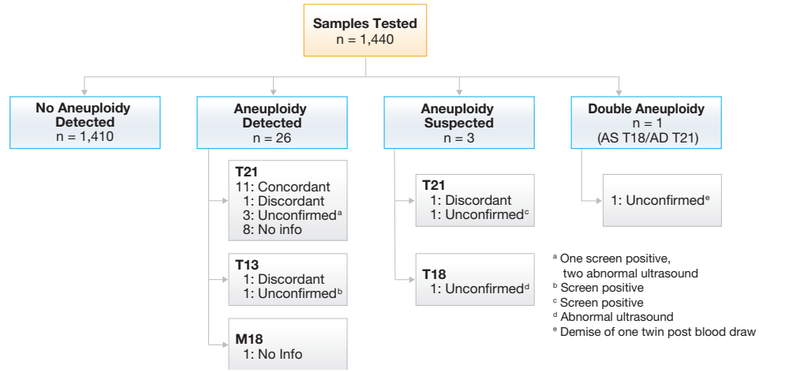
	Current study (n = 1,456)	Clinical Experience with singletons[1] (n = 34.860)
Maternal Age (years)		
Mean	35.9	35.44
Min - Max	18.3 – 53.5	13.8 – 53.2
Gestational Age (weeks)		
Mean	13.94	14.2
Min - Max	9[a] - 36	6[a] - 38
Trimester, n (%)		
First (up to 13 weeks)	921 (63.3)	21,533 (61.9)
Second (14 – 27 weeks)	519 (35.6)	12,773 (36.7)
Third (28 – 40 weeks)	16 (1.1)	470 (1.3)

[a] Samples received from patients under 10 weeks of gestational age were cancelled

Table 3. Clinical Laboratory Experience with Twin Gestations

Total Reported	1440
No Aneuploidy Detected	1410 (97.9%)
Aneuploidy Detected/Suspected	30 (2.1%)
Presence of Y Ordered	1318 (91.5%)
Presence of Y Detected	930 (70.6%)
Presence of Y Not Detected	388 (29.4%)

Figure 1 . Clinical Laboratory Experience with Twin Gestations*



**Majority of "unconfirmed" and "no info" outcomes are from non-US sites*

Summary

- An average turn-around time was 3.1 business days
- Mean maternal age is similar in twins and singletons, although the threshold for being considered of advanced maternal age is lower in twin gestations.
- Mean gestational age is significantly earlier in twin pregnancies compared to singletons ($p < 0.004$) and could reflect the trend toward earlier gestational age observed since test launch
- Ratio of twin pregnancies with no Y detected (29.4%) to ratio of twin pregnancies with Y detected (70.6%) nears the 1:2 expected ratio
- Cancellation rate (0.07%) due to technical reasons is similar in twin gestations to singletons (0.1%)
- No false negatives reported

Conclusions

- NIPT in twin gestations performs similarly to expected in singleton pregnancies, with no known false negative results.
- This study demonstrates that the verifi® test algorithm for twin gestations shows excellent test metrics in a clinical lab setting.
- Test metrics in twin gestations is similar to those in singletons[1] (Table 1 and 2), providing further support for the utility of NIPT in twin pregnancies.
- Continued efforts to obtain pregnancy outcomes in twin gestations are needed to better define clinical test performance of NIPT in this population.

References

1. Bhatt S, Parsa S, Snyder H, Taneja P, Halks-Miller M, Seltzer W, DeFeo E. Clinical Laboratory Experience with Noninvasive Prenatal Testing: Update on Clinically Relevant Metrics. ISPD 2014 poster.