

First Trimester Interlaboratory Comparison Program

Distribution 2008 FT-B



Sponsored by:
Department of Pathology and Laboratory Medicine
Women & Infants Hospital
Providence, RI

INTRODUCTION

Explanation of Data Listing and Analysis

Reading the Data Listing: The five page data listing (attached) contains a summary of reported results for all participants, with each page summarizing one specimen. Your lab ID is listed at the beginning of the row with your results. Missing data (blanks) are likely due to participants who are manufacturers rather than screening labs, or to laboratories that are not yet offering screening services. Missing data may also result because some laboratories do not measure 'total or intact hCG' but instead measure another marker (e.g., free beta hCG). Outliers for gestational age (or maternal age) are identified as those outside +/- 0.2 weeks (or years) of the correct answer. For the assay results (in mass units or MoM) and Down syndrome risks, outliers are defined as being outside of +/- 2 trimmed standard deviations, after accounting for rounding. A logarithmic transformation is used for the analysis of Down syndrome risk results.

Conversion of reported risks to first trimester risks. Almost all laboratories report first trimester risks, but some laboratories report second trimester or term risks. If the reported risks are not first trimester, then these risks are displayed in the column labeled "Report" under the "Down S Risk (1:n)" heading. To allow all risks to be evaluated by a single statistic, second trimester risks are converted to first trimester risks using the factor 0.74. This accounts for fetal loss between the first and second trimesters (43% from first trimester to term, and 23% from second trimester to term). For example, if the second trimester risk is 1:1000, the first trimester risk is $1:1000 \times 0.74$, or 1:740. Term risks are converted to first trimester risks by multiplying by 0.57.

Greater than and less than risks: Risks that are reported as less than (<) or greater than (>) are displayed in the "Report" column under the "Down S Risk (1:n)" column. These risks are listed as the actual numeric risk in the "1st trim" column and are included in the final calculation of the consensus risk.

RESULTS

FT-06: Laboratories were asked to calculate an NT MoM value, given a CRL of 45 mm (~ 11.6 weeks' gestation) and an NT value of 1.15 mm for the sonographer HFW. Participants were previously given a series of NT/CRL measurements for this sonographer and had already calculated a sonographer-specific median equation (sent again in this distribution for those who may need to recompute the medians). Participants may, or may not, have used those medians to calculate their MoM value, depending on existing laboratory protocols. The expectation is that the resulting MoM values reported by laboratories that use sonographer-specific medians should be similar. We calculated the median equation for sonographer HFW to be: median NT = $10^{(-0.209+0.005 \cdot \text{CRL})}$ using the EXCEL calculator supplied to participants. This equation yields an expected median NT value of 1.04 mm for a CRL of 45 mm, and, therefore, a MoM value of 1.11 (1.15/1.04). The consensus NT MoM (calculated as the median value) value was 1.11, in agreement with the consensus. Two results were on the low side (0.97 and 0.98 MoM), and another reported an outlying value (0.89 MoM). Some labs may not use the calculator supplied by the ICP, but instead use internal or commercial software that may yield a slightly different set of medians. Overall, the results indicate that laboratories can derive a common median equation, given a set of paired sonographer NT/CRL values, and can use those results to provide clinical interpretations.

A CRL was provided for this sample, requiring each lab to estimate gestational age. This was done to assess the variability in assigning gestational age by participating laboratories. As-

signed gestational ages for FT-06 ranged from 11.0 to 11.3 weeks. As pointed out previously, differences reflect the 'CRL to decimal weeks' equation selected by laboratories. The Supplemental Question in the 2005 FT-C report addresses this issue (accessible at <http://www.ipmms.org/ICP/FT-C%202005%20Final%20Report.pdf>), and includes a review of equations in common use. It is recommended that participants review this exercise if there are questions.

The CVs of the PAPP-A values and MoMs were higher than for the corresponding hCG measurements, in line with typical performance. However, the CV of log risk was very good (4%). All labs interpreted the specimen as screen negative.

FT-07: This specimen is a pool of sera from 12 week pregnancies, and should, therefore, more accurately reflect actual between-lab and between-kit differences. The CV for the PAPP-A value (14%) is slightly better than typical manufactured samples. However, the CV of the MoM (20%) is similar to other samples. The CVs for hCG were also typical of other samples. All labs reported the specimen as screen negative, and the CV of the log risk was very good (5%). One lab that offers sequential screening checked two answers (US/counsel for amnio and Collect new sample and retest) for the recommended action, and this has been listed as Other in the report (also true for some subsequent specimens).

FT-08: THE CVs of risk for both value and MoM for both markers were in line with typical performance. The high CV of log risk (22%) reflects the fact that most laboratories reported risks with only one digit of precision (e.g., 1:7).

FT-09: This specimen was targeted to have a high PAPP-A value, and the trimmed mean consensus value of 7.55 mIU/mL was consistent with the target. Overall, the results were good, with a CV of 12%, with one outlying value (4.2 mIU/mL). The elevated consensus PAPP-A MoM of 3.25, the consensus hCG MoM of 1.24, and the low NT MoM of 0.91, along with a maternal age of 38.1 years would be expected to yield a low risk. The consensus risk was 1:4940, with a low CV of 4% for log risk. However, three risks were considered to be outliers; two were low and one was high. All labs called the specimen screen negative and all but one recommended no further action.

FT-10: This sample was targeted to have a low PAPP-A and a high hCG, and the consensus MoM values were in line with expectation (0.41 and 3.55 MoM, respectively). Accordingly, the risk was high (1:34), and all labs called the specimen screen positive.

Dimeric inhibin-A (DIA)

First trimester DIA measurements were reported by four participants (Table 1). All reported using the same method (Di-0). The following table provides the reported DIA values and MoM levels for each of the five samples. Included also are the DIA likelihood ratios (LR) in the context of the other markers. Overall, the laboratories reported reasonably equivalent DIA values, MoM levels and likelihood ratios, with some indication that Laboratory A has higher DIA values and MoM levels.

Table 1. Dimeric Inhibin-A measurements for FT-B 2008

Sample Number	Laboratory	Value	MoM	DS Risk (1:n)	DIA LR ¹
FT-06	A	133.0	0.41	<10000	<1.0
	B	117.0	0.39	<10000	<1.0
	C	121.0	0.31	27500	0.47
	D	128.5	0.35	34100	0.38
FT-07	A	269.0	1.21	3400	0.91
	B	240.0	1.17	6400	0.61
	C	218.1	0.81	19300	0.20
	D	259.7	1.06	6720	0.43
FT-08	A	165	0.58	20	0.50
	B	156	0.64	8	0.50
	C	154.2	0.48	16	0.38
	D	157	0.54	8	0.63
FT-09	A	201.0	0.49	4600	1.11
	B	191.0	0.77	2200	0.39
	C	206.3	0.59	21800	0.24
	D	216.3	0.72	35600	4.19
FT-10	A	6020	19.73	16	2.19
	B	6140	21.90	6	3.00
	C	4624	12.71	4	4.50
	D	6714	22.41	5	9.40

¹ For each participant, the DIA LR is computed by dividing the reported risk for NT, PAPP-A and hCG by the risk that also includes DIA measurements. If blanks are shown, the likelihood ratio cannot be reliably determined, usually because one, or both, of the reported risks are very high (e.g., >1:10) or very low (e.g., <1:10,000).

Supplemental Exercise: Evaluation of Wild Card specimens

Two wild card specimens were included in the 2008 FT-B distribution to evaluate a new approach for manufacturing specimens. This approach will also allow us to offer the option of free beta hCG measurements as a routine in the near future. One sample was labeled PAPP-A/hCG WC and was to be assayed by those labs using hCG. The second sample was labeled PAPP-A/free beta WC for those laboratories using free beta hCG. All were to measure PAPP-A in both samples, as the concentrations were targeted to be identical. The hCG and free beta hCG concentrations were targeted for the first trimester. Table 2 summarizes the results.

Table 2. FT-B 2008 wild card results

PAPP-A Method	PAPP-A (mIU/mL) in specimen		hCG (IU/mL) ²	Free Beta (mIU/mL)
	with hCG	with free beta		
DSL	1.48	1.35	105.0	
	1.00	1.00	78.3	
	1.02	0.98	91.7	
	1.26	NR	NR	
	0.80	0.90	89.9	
	1.39	1.07	121.2	
	1.01	1.00	103.4	
	1.18	NR	94.1	
	1.04	0.98	109.0	
	1.34	1.24	107.9	
	1.54	1.39	106.4	
	1.20	1.20	96.6	
	0.92	0.95	110.2	
		2.40¹	155.5¹	
	Average	1.18	1.11	
SD	0.21	0.17		
CV	18%	15%		
DPC	4.41	4.23	94.0	
	3.97	3.64	102.0	
	3.51	3.7	77.9	
	Average	3.88	3.60	
SD	0.37	0.29		
CV	16%	13%		
PE	1.26	1.29	94.8	51.1
	1.22	1.19	96.2	52.7
	1.28	1.23	96.7	50.6
	Average	1.25	1.24	98.6²
SD	0.03	0.05	10.9	1.1
CV	2.4%	4.1%	11%	2.1%

¹ Values excluded from calculations

² hCG results not stratified by method

The PAPP-A values for both specimens was targeted to have a nominal concentration of 1.0 to 1.5 mIU/mL. The results for the DSL and PE kits were close to expectation, and the overall average for the two methods was similar (about 1.2 IU/mL). However, the DPC users had consistent results that were more than 2 times higher (about 3.7 IU/mL). Such a discrepancy does not exist when pools of patient sera are used (e.g., specimen FT-07 2008). This indicates that the manufacturing method we used for the wild card specimens is not suitable the ICP. We are exploring an alternative approach to use for FT-C.

The overall results for hCG were close to the target value of 100 IU/mL, and the between lab CV is similar to that observed for samples made from pools of patient material (11%). The average free beta result of 51.5 mIU/mL met the expected target of 50 mIU/mL, indicating that the use of recombinant material is likely to be suitable.

We wish to thank all of you for assisting us in evaluating different methods for manufacturing specimens that are stable, yield results found with actual patient sera and can be appropriately targeted. This is proving to be a challenge, but not an insurmountable one.

Interpretive Questions – Integrated Screening for Down Syndrome

- 1. Does your laboratory provide clinical Down syndrome screening services?** Twenty laboratories reported that they screen for Down syndrome. Two did not respond to the question.
- 2. Does your laboratory perform integrated risk interpretations?** Among the 20 responding laboratories, 13 reported that they offer integrated screening as part of a formal screening program, one lab upon request, and the remaining six reported that they do not offer integrated screening. All laboratories used the same 'trimester of risk' for their quadruple and integrated Down syndrome risks (second trimester or term risks). All 13 laboratories could provide (including NT) integrated risks using all four second trimester markers (AFP, uE3, hCG and DIA).
- 3. Report the Down syndrome risks from FP-12 (CAP FP-B 2008 Survey). Report integrated risks using FT-09 results (with modifications to the draw date).** The consensus risk (median value) for the laboratories reporting integrated results for quadruple markers was **1:237**. Given the PAPP-A consensus MoM of **2.47**, one would expect each laboratory to calculate a serum integrated test risk that is lower than its quad risk. The median risk for the serum integrated test was **1:967**. This reduction can be expressed as a likelihood ratio (LR), obtained by dividing the quad risk by the serum integrated risk (column 4 in Table 3). The consensus LR for these two risks is 0.24 (237/967), consistent with the LR for all labs (0.23) reported in the table. One laboratory reported a LR greater than one, indicating an increased risk (1.73), which is an outlier. The consensus risk (median) for the full integrated test is 1:2680, yielding a consensus LR of 0.09 (237/2680) when divided into the quad risk, again consistent with the consensus LR of 0.09 in Table 3. Column 6 in the table displays the likelihood ratio that would result if the NT measurement were unavailable, and only a serum integrated test could be reported out. The risk would increase from 1:2680 to 1:967, yielding a likelihood ratio of 3.32. One lab found a decrease in risk, yielding an outlying ratio of 0.47. One cause for outlying risks is the use of different parameter sets for the quad test versus integrated testing. In a future distribution, we plan to expand the information requested to further explore the sources of outlying risks.

Table 3. Comparison of quadruple risks to integrated risks.

Down syndrome risk (1:n)			Likelihood ratio ¹		
Quadruple (Q) (FP-12)	Full Integrated (FI) (FP-12 & FT-09)	Serum Inte- grated (SI) (FP-12 & FT-09)	Q/SI	Q/FI	FI/SI
18	106	224	0.08	0.17	0.47
78	1700	870	0.09	0.05	1.95
110	1300	620	0.18	0.08	2.10
115	923	271	0.42	0.12	3.41
120	1000	400	0.30	0.12	2.50
190	210	110	1.73	0.90	1.91
234	20000	4100	0.06	0.01	4.88
240	2600	660	0.36	0.09	3.94
275	2760	908	0.30	0.10	3.04
295	3400	1200	0.25	0.09	2.83
370	3400	790	0.47	0.11	4.30
470	6500	1300	0.36	0.07	5.00
600	8100	1700	0.35	0.07	4.76
620	9100	1700	0.36	0.07	5.35
237	2680	967	← Median reported risk		

outlier	1.73		
Trimmed LR	0.23	0.01, 0.90	0.47
log SD of LR	0.30	0.09	3.32
CV	48%	0.15	0.16
Mean - 2sd	0.06	14%	31%
low	0.06	0.05	1.60
high	1.73	0.01	1.95
Mean + 2sd	0.91	0.90	5.35

¹ Derived by dividing the associated Down syndrome risks.

George J. Knight, Ph.D.
Jacob A. Canick, Ph.D.

Glenn E. Palomaki, B.S.
Geraldyn M. Messerlian, Ph.D.

(207) 657-7888
(401) 453-7650

Department of Pathology and Laboratory Medicine
Women & Infants Hospital
Providence, Rhode Island