NEW YORK
state department of
HEALTH

Nirav R. Shah, M.D., M.P.H.
Commissioner

Sue Kelly
Executive Deputy Commissioner

New York State FEDM – Proficiency Testing Program

TO: Laboratory Directors

CATEGORY: Fetal Defect Markers (FEDM)

MAILOUT: September 13, 2011

FROM: Dr. G.J. Mizejewski, Director of FEDM Program

DUE DATE: September 28, 2011

Samples:
There are five (5) vials labeled **MS271** to **MS275**, each containing various predetermined amounts of alpha-fetoprotein (**AFP**), human chorionic gonadotropin (**hCG**), unconjugated estriol (**uE3**) and Dimeric **Inhibin A**. Also, five additional vials (AF 271 to AF 275) containing AFP in amniotic fluid have also been included. In addition, five extra vials **FT 271 to FT 275** containing human chorionic gonadotropin (**hCG**) and **PAPP-A** are added for ***optional*** testing. Please note that you do not have an option if you offer First Trimester and or Integrated Testing but the results of **FT 271 to FT 275** will ***not be graded***. Please analyze for all of those markers tested in your laboratory the same way as you would with a patient sample. If your lab is also measuring Amniotic fluid AFP, you are also required to measure those samples provided. Maternal serum samples are in human-derived serum base, sterile filtered and dispensed. Please **keep refrigerated** until use, but do ***not*** freeze. Before analyzing, make sure samples are mixed completely.

Reporting of Results:
All laboratories **must** submit their proficiency testing results electronically through the electronic proficiency testing reporting system (**EPTRS**) on the Department's Health Commerce System (HCS). The HCS is a secure website and requires all users to obtain an account ID in order to access the HCS and EPTRS application. The portal's URL is https://commerce.health.state.ny.us. Questions regarding the entry and submission of proficiency test results or the account application process can be directed to clepeptrs@health.state.ny.us. If your laboratory does not have an HCS account, you must request one as soon as possible before the next PT event by contacting the Clinical Laboratory Evaluation Program at 518-486-5410. Also, please also **see attached September 2011 bulletin**.

For help with logins, password problems and reactivating HCS accounts, contact the Commerce Account Management Unit (CAMU) at (866) 529-1890.

Results must be reported for all 5 Maternal Sera and/or Amniotic fluid samples; otherwise a zero grade will be applied to the missing data. Please enter your mass unit results in the spaces provided with one or two decimals accordingly. If a result exceeds your analytical range, indicate this with a "less than (<)"or "greater than (>)" sign if similar results from patient samples are reported in the same manner. If such samples are routinely retested after dilution, you may do so provided the result is identified accordingly. Select the instrument and reagent/kit used for each analyte using the drop-down menus. Please note that the risk factor and further action (not graded) for each of the samples has also been placed in the EPTRS. All applicable fields must be completed. Missing entries will result in a failing grade for the missing results.

If CLEP is contacted for permission to submit results via paper, this request may be approved under extenuating circumstances. However, the lack of active HCS accounts, the lack of submission roles, or the lack of Internet access will not excuse a laboratory from having to submit results electronically. Without such approval, mailed or faxed proficiency test results will not be accepted. Note that such approvals will not be given on the due date! If you have any questions, please call Ms. Helen Ling at (518) 474-0036.

Please Turn Over

Special Instructions:
In order to achieve uniformity among our labs in reporting gestational age results, please report gestational week in "**decimal weeks (weeks + day/7)**" for the maternal serum samples.

Example: 18,3 weeks in the Ultrasound dating means 18 weeks + 3 days or 18.4 weeks (18 weeks + 3/7 weeks) ***not*** ~~**18.3**~~, i.e. **18.4 should be reported**

Note: We recommend the use of LMP (ultrasound dating when available) in calculating the gestational age, please note that the use of EDD is not an accepted standard of patient care.

Caution:
All human derived specimens should be handled as biohazard materials using Universal Precautions.

Only extra correspondence and information about **new kits** may be mailed to**:**

Fetal Defect Markers Proficiency Testing c/o Helen Ling
Wadsworth Center
Empire State Plaza, Room E610
PO BOX 509
Albany, NY 12201-0509

Please let us know immediately if you do not receive the samples in satisfactory condition by calling Ms. Helen Ling at (518) 474-0036.

DUE DATE: Results must be submitted electronically before 11:59 PM of September 28, 2011.
*Test results will not be evaluated if the results are **submitted** after the due date and a Failing Grade will be assigned.*

The next Proficiency Test mail-out for 2012 has been tentatively scheduled for:

Ship-out date	**Due date**
January 24, 2012	February 8, 2012
May 8, 2012	May 23, 2012
September 11, 2012	September 26, 2012

Demographic Data:

Specimen	Maternal Date of Birth	Race[1] W,B,H,A	Maternal Weight (lbs)	IDD[2] Presence	Gravida	Parity	LMP[3]	Draw Date
MS 271	**9/16/1982**	**W**	**150**	**None**	**1**	**0**	**5/27/2011**	**9/9/2011**
MS 272	9/16/1981	H	200	None	2	1	5/13/2011	9/9/2011
MS 273	9/15/1986	W	145	None	1	0	5/6/2011	9/9/2011
MS 274	9/14/1990	A	120	None	3	2	4/29/2011	9/9/2011
MS 275	**9/14/1988**	**W**	**155**	**None**	**3**	**1**	**4/22/2011**	**9/9/2011**

Specimen	GA[4]
AF 271	**15.0**
AF 272	19.0
AF 273	20.0
AF 274	20.9
AF 275	**20.0**

***Note: MS271 and MS275** are the serum sample matched to the amniotic fluid sample **AF271 and AF275, respectively**. (Dating by ultrasound)

[1]Race: W = White, not of Hispanic origin; B = Black, not of Hispanic origin; H = Hispanic; A = Asian

[2]IDD = Insulin-Dependent Diabetic

[3]LMP = Last Menstrual Period

[4]GA = Gestational Age in Decimal Weeks

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Fetal Defect Marker Proficiency Test Mailout[1]
September 2011

Dear Laboratory Director,
Below you will find a summary and critique of the Proficiency Testing mail-out from September 13, 2011, for Fetal Defect Markers, which included samples for first and second trimester screening, as well as amniotic fluids. Your laboratory's results and grades are printed on a separate sheet; also included are the grades from the previous two PT events. These will be mailed to you separately. Please review and sign your evaluation. Retain the signed evaluation in your files. You will need it for your next laboratory survey to demonstrate participation in the NYSPT program.

I. Graded Results Section: Table 1: Second Trimester Maternal Serum: Summary of All Lab Results

Samples *N = 27	**Sample #**	**MS 271**	**MS 272**	**MS 273**	**MS 274**	**MS 275**
	Gestational Age (weeks)	15.0	17.0	18.0	19.0	20.0
Maternal Race	Ethnic Group	White	Hispanic	White	Asian	White
Maternal Weight	Pounds (lbs)	150	200	145	120	155
Maternal Age	Years	29	30	25	21	23
Alpha-Fetoprotein (AFP)	Mean ng/ml ± Std. Dev.	14.20 ± 1.20	37.51 ± 3.00	115.00 ± 8.91	57.40 ± 5.41	172.11 ± 17.10
	MOM ± Std. Dev.	0.49 ± 0.05	1.18 ± 0.11	2.55 ± 0.22	0.97 ± 0.12	2.97 ± 0.32
Unconjugated Estriol (uE3)	Mean ng/ml ± Std. Dev.	0.32 ± 0.04	0.94 ± 0.08	1.06 ± 0.07	1.26 ± 0.12	1.42 ± 0.15
	MOM ± Std. Dev.	0.58 ± 0.18	1.14 ± 0.32	0.93 ± 0.21	0.86 ± 0.24	0.86 ± 0.24
human Chorionic Gonadotrophin (hCG)	Mean IU/ml ± Std. Dev.	64.40 ± 8.35	19.93 ± 1.91	17.77 ± 1.77	15.29 ± 1.45	14.10 ± 1.25
	MOM ± Std. Dev.	1.71 ± 0.23	1.00 ± 0.10	0.86 ± 0.09	0.74 ± 0.09	0.85 ± 0.09
Dimeric Inhibin-A (DIA)	Mean pg/ml ± Std. Dev.	305.82 ± 36.65	168.30 ± 20.24	148.50 ± 17.82	212.79 ± 25.61	245.15 ± 31.16
	MOM ± Std. Dev.	1.63 ± 0.24	1.13 ± 0.17	0.89 ± 0.08	1.11 ± 0.18	1.29 ± 0.19
Neural Tube Screen (Positive, Negative) Percent	Pos. (+) or Neg. (-)	(-) (100%)	(-) (100%)	(+) (66%)	(-) (100%)	(+) (100%)
	Further Action G,U,A	NFA	NFA	G = 56% U = 56% A = 41%	NFA	G = 85% U = 93% A = 78%
	NTD Risk 1 in	5,750	6,750	295	6,840	80
Trisomy-21 Screen (Positive, Negative) Percent 1. Triple test	Pos. (+) or Neg. (-)	(+) (100%)	(-) (100%)	(-) (100%)	(-) (100%)	(-) (100%)
	Recommended Action**	G = 86% U = 64% A = 79%	NFA	NFA	NFA	NFA
	Risk Est. 1 in	86	4,479	9,496	8,100	8,157
2. Quad Test	Pos. (+) or Neg. (-)	(+) (96%)	(-) (100%)	(-) (100%)	(-) (100%)	(-) (100%)
	Recommended Action **	G = 85% U = 67% A = 85%	NFA	NFA	NFA	NFA
	Risk Est. 1 in	35	6,200	9,000	10,000	8,398
Trisomy-18 Screen (Positive, Negative) Percent	Pos. (+) or Neg. (-)	(-) (100%)	(-) (100%)	(-) (100%)	(-) (100%)	(-) (100%)
	Recommended Action**	NFA	NFA	NFA	NFA	NFA
	Risk Est. 1 in	4,132	52,500	48,550	18,150	27,100

*N = total numbers may vary since some labs do not test all analytes. The values represent the all-lab consensus based on the arithmetic mean ± Std. Dev.;
(B) = borderline positive or negative, risk reflects central tendency (Median number for NTD/Down positive or negative/borderline screen). NFA = no further action; FA = further action; G = genetic counseling; U = ultrasound, and A = amniocentesis.
**This percentage is normalized to labs requesting further action. ‡ Insulin Dependent Diabetic pregnancy.

[1]The use of brand and/or trade names in this report does not constitute an endorsement of the products on the part of the Wadsworth Center or the New York State Department of Health.

1) Second Trimester Maternal Serum Analytes:

A. Narrative Evaluation of Second Trimester Screening Results:

N = 27 all-lab Consensus Values.

Sample #	Summary Comments (Mock specimens):
MS 271 Wk 15.0	This specimen was obtained from a 29 year old White woman (Gravida = 1, Parity = 0) in her 15th week of gestation with a body weight of 150 lbs. She had a family (sibling) history of pregnancy complications. Her sample screened negative for NTD; however, her aneuploidy screen was positive for Trisomy-21 (96% Quad, 100% Triple) on the basis of low AFP and uE3, and moderately elevated hCG and Inhibin-A levels. Recommendations for further action from labs performing the T21 quad screen were: genetic counseling, 85%, ultrasound, 67% and amniocentesis, 85%; while the triple tests were: genetic counseling, 86%; ultrasound, 64% and amniocentesis, 79%. Specimen MS271 resulted in a negative T18 screen in 100% of the participating labs. This sample was paired to an amniotic fluid specimen, which also had a low AFAFP level (MOM = 0.53).
MS 272 Wk 17.0	This specimen was obtained from a 30 year old Hispanic woman (Gravida = 2, Parity = 1) in her 17th week gestation with a body weight of 200 lbs. She had no family history of pregnancy complications. To date, her pregnancy appeared to follow a favorable course of gestation, and her specimen resulted in a negative screen for NTD with a body weight correction indicated. The labs were also in agreement that both Trisomy screens were negative. Specimen MS272 was not paired with an amniotic fluid sample.
MS 273 Wk 18.0	This specimen was obtained from a 25 year old White woman (Gravida = 1, Parity = 0) in her 18th week of gestation with a body weight of 145 lbs. She had a family (sibling) history of accelerated body growth and advanced bone age. Her sample screened borderline positive for NTD; however, her aneuploidy screen was negative for Trisomy-21 (100%). This sample was not paired to an amniotic fluid specimen. Please see critique for further discussion of MS273.
MS 274 Wk 19.0	This specimen was obtained from a 21 year old Asian woman (Gravida = 3, Parity = 2) in her 19th week of gestation with a body weight of 120 lbs. A race correction may be indicated. She had no personal history of pregnancy loss. Her specimen was negative for NTD and for both Trisomies with all labs in agreement. Thus, no recommendations for further action were noted. This specimen had no amniotic fluid counterpart.
MS 275 Wk 20.0	This specimen was obtained from a 23 year old white Woman (Gravida = 3, Parity = 1) in her 20th week of gestation with a body weight of 155 lbs. She had a family (sibling) history of reproductive complications. Her sample screened positive for NTD, and her aneuploidy screens were negative for both Trisomy-18 and Trisomy-21. The MS275 sample was paired to an amniotic fluid specimen, which was elevated (AFAFP MOM = 2.87). Please see Critique below for further discussion of samples MS275 and AF275.

Notice of Gravida/Parity Clarification for Present and Future Mail outs;

Instructional Note:

This notice regards the demographic data provided for the mock patients in the FEDM program. For the sake of this program, it will be understood that gravida indicates the pregnant status of a woman and parity is the state of having given birth to a completed term infant or infants. Thus, a gravida = n, indicates number (n) of times pregnant including the present one; a gravida = 2 indicates that the women was pregnant once before in addition to her present pregnancy. Parity = 1 indicates the patient already has one child; however, multiple birth is also considered as a single parity.

Example: A woman of gravida = 3, parity = 2 indicates that the pregnant woman has been pregnant twice before, and has two children.

2) AMNIOTIC FLUID AFP (NTD-analysis):

N=27; all-lab Consensus Values

Sample#	Values	Summary Comments:
AF 271 Wk 15.0	AFP = 9.29 ± 1.50 µg/ml MOM = 0.53 ± 0.06	The AF271 sample was targeted for a low AFAFP value in the lower gestational age range. All labs called AF271 a non-elevated specimen for NTD. This AFAFP sample was matched to a maternal serum specimen which was also low (MOM = 0.49).
AF 272 Wk 19.0	AFP = 4.56 ± 0.57 µg/ml MOM = 0.57 ± 0.05	The AF272 sample was targeted for a negative NTD screen for AFAFP in the upper gestational age screening range. All labs categorized this as an NTD screen negative specimen. This sample was not matched to a maternal serum specimen.
AF 273 Wk 20.0	AFP = 7.14 ± 1.29 µg/ml MOM = 1.09 ± 0.15	The AF273 sample was targeted for a normal AFAFP value in the upper gestational age range. All labs called AF273 a normal MOM AFAFP specimen. This AFAFP sample was not matched to a maternal serum specimen.
AF 274 Wk 20.9	AFP = 6.00 ± 0.88 µg/ml MOM = 1.04 ± 0.14	The AF274 sample was targeted as an NTD negative screen in the upper gestational age screening range. All labs categorized AF274 as a negative NTD screen specimen. This specimen had no maternal serum counterpart.
AF 275 Wk 20.0	AFP = 18.71 ± 2.88 µg/ml MOM = 2.87 ± 0.32	The AF275 sample was targeted for a screen positive AFAFP value in the upper gestational age range. All labs reported this specimen as a screen positive AFAFP value. The AF275 specimen was paired with maternal serum sample MS275, which was positive (MOM = 2.97). Please see Critique below for further discussion of samples MS275 and AF275.

II. Non-Graded Results Section:

Table 2: First Trimester Maternal Serum all-lab Results

Samples *N = 17	**Sample #**	**FT 271**	**FT 272**	**FT 273**	**FT 274**	**FT 275**
	Gestational Age (weeks)	11.5	11.9	11.2	12.4	13.1
Maternal Race	Ethnic Group	Hispanic	White	Asian	Hispanic	White
Maternal Weight	Pounds (lbs)	160	150	100	140	130
Maternal Age	Years	29	25	21	26	19
Nuchal Translucency (NT)-Associated Measurements	Crown Rump Length (mm)	48	53	45	59	69
	NT Thickness (mm)	1.08	2.90	1.10	1.40	1.55
	NT – MOM ± Std. Dev.	0.89 ± 0.05	2.17 ± 0.16	0.95 ± 0.06	0.95 ± 0.06	0.93 ± 0.06
Human Chorionic Gonadotrophin (hCG) Total	Mean IU/mL ± Std. Dev.	70.09 ± 9.08	148.04 ± 27.36	69.56 ± 9.59	59.49 ± 6.88	56.62 ± 5.63
	MOM ± Std. Dev.	0.95 ± 0.11	1.99 ± 0.23	0.72 ± 0.12	0.83 ± 0.08	0.82 ± 0.10
Pregnancy-Associated Plasma Protein–A (PAPP-A)	Mean ng/mL*** ± Std. Dev.	1629.23 ± 312.86	798.22 ± 103.63	1407.35 ± 171.21	2055.52 ± 412.36	2307.52 ± 346.91
	MOM ± Std. Dev.	3.24 ± 1.60	1.36 ± 0.68	2.05 ± 1.04	2.51 ± 1.22	2.01 ± 1.03
Trisomy-21 Screen (Positive, Negative) Percent	Pos (+) or Neg. (-)	(-) (100%)	(+) (93%)	(-) (100%)	(-) (100%)	(-) (100%)
	Recommended Action NFA**	NFA	G = 93% U = 33% A = 40% C = 53%	NFA	NFA	NFA
	Risk Estimate	19,000	1 in 38	23,900	21,000	24,000
Trisomy-18 Screen (Positive, Negative) Percent	Pos (+) or Neg. (-)	(-) (100%)	(-) (100%)	(-) (100%)	(-) (80%)	(-) (100%)
	Recommended Action	NFA	NFA	NFA	NFA	NFA
	Risk Estimate	119,000	3,510	119,000	119,000	119,000

*N = total numbers may vary since some labs do not test all analytes. (B) = borderline negative or positive; NFA = no further action; G = genetic counseling; U = ultrasound; C = chorionic villus sampling; N = number of labs participating; FT = First Trimester.
**This percentage is normalized to labs requesting further action.
***Results from methods that give mIU/ml were converted to ng/ml as described in section D.1 below.

1) First Trimester Maternal Sera Only:

B. Narrative Evaluation of First Trimester Screening Results:
N = 17 all-lab Consensus Values.

Sample#	Summary Comments:
FT 271 Wk 11.5	This specimen was obtained from a 29 year old Hispanic woman of normal body weight (160 lbs.). Her gestational age at the time of screening was 11.5 weeks. She had no prior history of pregnancy complications and/or adverse outcomes. This FT specimen was screen negative with all testing labs in agreement. The FT271 risk estimate for Trisomy-21 was 1 in 19,000, while the Trisomy-18 risk was 1 in 119,000.
FT 272 Wk 11.9	This specimen was procured from a 25 year old White woman of average body weight (150 lbs.). Her gestational age at the time of screening was 11.9 weeks. She had a prior family history of pregnancy complications and/or adverse outcomes. This FT specimen was screen positive for Trisomy-21 and all but one of testing labs were in agreement (see Critique). The FT272 risk estimate for Trisomy-21 was 1 in 38, while the Trisomy-18 risk was 1 in 3,510.
FT 273 Wk 11.2	This specimen was obtained from a 21 year old Asian woman of low body weight (100 lbs). Her gestational age at the time of screening was 11.2 weeks. She had no prior history of pregnancy complications and/or adverse outcomes. This FT specimen was screen negative and all testing labs were in agreement. The FT273 risk estimate for Trisomy-21 was 1 in 23,900, while the Trisomy-18 risk was 1 in 119,000.
FT 274 Wk 12.4	This specimen was procured from a 26 year old Hispanic woman of average body weight (140 lbs.). Her gestational age at the time of screening was 12.4 weeks. She had no prior history of pregnancy complications and/or adverse outcomes. This FT specimen was screen negative for Trisomy-21 and all testing Labs were in agreement (see Critique). The FT274 risk estimate for Trisomy-21 was 1 in 21,000, while the Trisomy-18 risk was 1 in 119,000.
FT 275 Wk 13.1	This specimen was procured from a 19 year old White woman with a body weight (130 lbs.). Her gestational age at the time of screening was 13.1 weeks. She reported no prior family history of pregnancy complications. This FT specimen was screen negative for Trisomy-21 and Trisomy-18. The Trisomy-21 risk estimate for FT275 was 1 in 24,000, and the Trisomy-18 risk was 1 in 119,000. All labs were in agreement with both screen assessments.

III. Critique and Commentary:

Critique:

A) Second Trimester Maternal Serum and Amniotic Fluid:

In general, the all-lab results were consistent with the targeted values for the NTD and the Trisomy Screens for risks, and outcomes. The Caucasian maternal serum sample **MS275** was targeted as positive specimen for NTD (Figs. 1 and 3) and was matched to an elevated **AF275** sample (Figs. 3 & 4; see discussion below). All labs (100%) agreed that specimen **MS275** was screen positive for NTD and negative for both Trisomy screens, and that AFP levels were elevated for amniotic fluid (see below). The **MS275** sample generated recommendations for further action. Sample **MS271** was obtained from a white woman with a prior family (sibling) history of pregnancy complications. The T21 MOM results for specimen **MS271** (MSAFP-MOM = 0.49, MSuE3-MOM = 0.58, MShCG-MOM = 1.71, DIA-MOM = 1.63) were consistent with a T21 positive screen; thus, almost all labs (100% triple and 96% quad) classified this specimen as T21 screen positive and recommended further action as follows. The T21-related recommended action for **MS271** triple screen was genetic counseling, 86%; ultrasound, 64%; and amniocentesis, 79%; while the quad test recommended action was genetic counseling, 85%; ultrasound, 67% and amniocentesis was 85%. The **MS271** sample produced a risk from the triple test risk of 1 in 86 and a quad test of 1 in 35. Two other specimens, **MS272**, and **MS274** produced negative screens for NTD, T21, and T18; corrections for body weight and race were suggested (see above). The **MS273** specimen, a special case involving elevated level of MSAFP, will be discussed below.

The **MS275** sample was screen positive for NTD and negative for T21 and T18; the amniotic fluid sample paired with this specimen was also elevated. Furthermore, the **AF275** sample was determined to have an elevated AFP value by all participating laboratories. Follow-up for **MS275** NTD screen was the following: genetic counseling, 85%; ultrasound, 93%; amniocentesis, 78%; and repeat sample, 4%. This mock patient had been referred to a tertiary care medical center for an amniocentesis due to a family history of pregnancy complications and poor outcomes in several extended and close family members. A maternal serum sample was obtained just prior to the amniocentesis; following amniocentesis, the post-procedure AF specimen (untainted by color) together with the MS sample was then analyzed at a tertiary care center. The final outcome in this mock patient showed that level-II diagnostic ultrasound showed the presence of a neural tube defect, and a diagnostic Ache band was present following gel electrophoresis.

The specimen **MS271** was designed to represent a positive screen for Down Syndrome with the typical MS profile of low MSAFP, low MSuE3, with elevated MShCG and MSDIA constituting the classical "quad test". With the addition of MS-DIA in second trimester screening, the detection rate in the literature has been found to increase to 75% (from 65%) while maintaining a 5% false positive rate (50). In the case of specimen **MS271**, the MS-DIA MOM value of 1.63 increased the T21 risk value of 1 in 86 (triple test) to a greater risk of 1 in 35 (quad test). This increased risk screen was exemplified by the "further actions" reported by the participating laboratories (see above paragraph). However, the all-lab positive T21 screen for **MS271** using the quad test was 96% compared to the triple test which was 100% positive screen due to individual lab risk cutoff values.

Since the **MS273** sample was screen positive for NTD, but negative for T-21, and T-18, the MSAFP in this specimen was of special interest. The follow-up actions for **MS273** were the following: genetic counseling, 56%; ultrasound, 56%; amniocentesis, 41%; and repeat, 15%. The **MS273** sample was determined to have an elevated MSAFP (MOM = 2.55) and normal MShCG (MOM = 0.86), uE3 (MOM = 0.93), and DIA (MOM = 0.89) values by all participating laboratories. This mock patient was then referred to a tertiary care medical center for a consultation due to a family history of accelerated body growth, advanced bone growth, and precocious puberty in several extended and close family members. The maternal serum sample had been sent to a prenatal biomarker screening and tertiary care center following the initial consultation, but amniocentesis had not yet been performed. At this time, **MS273** patient was not a suspected candidate for Hereditary Persistence of AFP, which is an autosomal genetic disorder. The lab amniocentesis and ultrasound results in showed a normal AFAFP levels and karyotype and level-II diagnostic sonography revealed no presence of NTD or trisomy-related defects; nor was any other structural or anatomic anomaly detected. The **MS273** sample with an unexplained elevated AFP was NTD screen positive with a normal amniotic AFP level. Due to the advanced body and bone growth and early puberty, an endocrinologist suggested that the patient and other family members of this patient undergo serum screening for AFP and other reproductive serum markers. Serum elevated AFP was found in multiple family members across three generations. The patient (proband) was then diagnosed as Hereditary Persistence of AFP.

Unexpected elevated level of MSAFP in NTD screening programs has always remained a cause for concern in the screening laboratory. Specimen **MS273** presents an interesting case in that it was designed to represent a rare benign autosomal dominant trait with complete penetrance referred to as Hereditary Persistence of alpha-fetoprotein (HP-AFP). The first clinical case was described in 1983 from an antenatal screening program for spina bifida (1, 2). A 38 year old Caucasian woman in her 21st week of pregnancy displayed highly raised levels of MSAFP. Ultrasound revealed a normal singleton fetus at the correct gestational age and amniotic fluid AFP levels were normal as in the present **MS273**. Her newborn baby was unremarkable at time of delivery with no visible anatomical malformations or clinical disabilities. Subsequently, 70 members of the family were tested and 21 members (including nine males) were found to have high levels of AFP in their serum. Since that time, nineteen families have been identified in the biomedical literature, some of which were coincident with testicular abnormalities in the males. Moreover, the inherited trait can be present in both male and female descendants within a given family line; however, the females rarely demonstrate any severe clinical disabilities or symptoms. Elevated AFP levels in adults may be present in a variety of conditions other than pregnancy, which can include liver and germ cell disorders, cirrhosis, hepatitis, malignancies, anemias, immunodeficiencies, and ataxia telangiectasias (3, 4). Thus, HP-AFP as a cause of persistently elevated AFP levels into adulthood can only be discerned by the testing of 1st and 2nd generation family members followed by molecular DNA studies to determine the basis of the trait. Such elevated AFP levels may be difficult to distinguish in patients being tested for malignancies, liver dysfunctions, and pregnancy.

As stated above in the adult non-pregnant population, AFP has long served as a biomarker for hepatocellular carcinomas, liver dysfunction, and non-seminomatous germ cell tumors (5, 12). Serial levels of serum AFP are monitored following tumor surgeries and these levels usually recede to normal which are less than 10 ng/ml. Patients with non-receding AFP values are presumed to have a recurrent or persistent disease and are usually treated with chemotherapy. This standard of care, however, does not take into account the presence of the rare disorder HP-AFP, which could manifest as a continued elevated AFP level. Such a case was reported in a 20 month old (2,659 ng/ml) male child who underwent surgery for a testicular yolk sac tumor and needlessly received multiple rounds of chemotherapy (6). The authors of this report proposed that in similar situations, parental levels of AFP should be assayed before deciding on chemotherapy based solely on mildly or moderately elevated AFP. The AFP blood test is a relatively simple and inexpensive test that could avoid unnecessary exposure to potentially toxic treatments. Several other cases in males with testicular seminoma (7), benign testicular cyst (11), testicular pain (8) and seminoma have been described (9, 10). In another instance, persistently elevated AFP levels were found in a healthy 43 year old man and subsequently found in three of his first degree relatives; these included two siblings, and one daughter of reproductive age (11). In a third case, a 39 year old man with a testicular fibroid nodule demonstrated HP-AFP as did 5 of 13 relatives within a 3 generation span (12). The reported AFP levels in these cases ranged from 18 to 198 ng/ml with no observed disease or functional abnormalities. Finally, HP-AFP was reported in a 42 year old man (AFP = 43 ng/ml) undergoing removal of a testicular tumor stage 1 Seminoma (8). AFP levels in his mother and sister ranged from 32 to 65 ng/ml. It should be noted that in non-HP-AFP patients, AFP is elevated only in cases of non-seminoma tumor, but not in seminoma tumors. Therefore, elevated AFP in a seminoma might be suggestive of HP-AFP.

To rule out liver involvement, lectins such as Concanavalin-A (Con-A), can be used to determine the tissue origin of AFP from the structure of the sugar chain on AFP. On Con-A columns, AFP filtration produces "binding" and "non-binding" fractions. If the fetal protein originates from a germ cell or gastrointestinal (GI) organ, a non-binding AFP form is found. However, a Con-A binding fraction indicates that AFP originated from the liver or a hepatoma. In effect, the Con-A binding test can differentiate yolk sac (GI) from liver tumors (13). In the mothers and adult sisters of the lectin-tested male patients with testicular dysfunction and tumor, the serum AFP values ranged from 32 to 65 ng/ml.

AFP belongs to the albuminoid gene family and all members are tandemly linked in the 4q subcentromeric region of the same DNA-strand and map to the locus 4q11 – 4q13 (14). A gene sequence analysis in the five-prime flanking region of the AFP gene obtained from a family with HP-AFP revealed a mutation in a potential enhancer element (G-to-A transition at position -119) called the Hepatocyte Nuclear Factor-1 Binding site (15). In a competitive gel retardation assay, the mutant sequence was found to bind Hepatocyte Nuclear Factor-1 (HNF-1) more tightly than did the wild type sequence. They also found that 5'-flanking sequences of the human AFP gene containing the G-to-A substitution directed a higher level of CAT expression in transfected human hepatoma cells than the wild type sequences. Their results emphasized the importance of the HNF-1 binding site on DNA in the developmental regulation of the human AFP gene (16).

A report of HP-AFP in a Spanish family concerned a 48 year old woman suffering from asthenia (fatigue and weakness), that also involved 8 of 16 family members in 3 generations (17). Molecular analysis of the woman and all affected members revealed the classical G to A substitution at an additional position -119 of the 5'- flanking region and was absent in all members showing normal AFP levels. The AFP levels in the first generation members ranged from 364 to 881 ng/ml, while those of the next generation were 240 to 583 ng/ml. Further molecular studies of HP-AFP from various ethnic groups involved a family of Bengali origin and one of Italian descendents (19). The Bengali family showed the previously reported distal mutated promoter G to A substitution at -119, while the Italian members exhibited a C to A, T (-65) and a C to A (-55) in the proximal HNF-1 binding region of the promoter. However, gel shift and transfection experiments failed to show any biological effect of the HNF-1 substitution associated with the C to A mutation but did so with the A to C mutation. Thus, at least one other mutation present near the HNF-1 binding sites of the AFP gene promoter did result in the HP-AFP phenotype. The C to T resulted only in formation of a CCAAT box.

In sole contrast to the above discussion, a large Taiwanese family demonstrating the HP-AFP trait did not exhibit the single nucleotide polymorphism (SNP) of previous studies involving the AFP promoter -199 G to A mutation. Seven members of this Asian family with the HP-AFP trait also failed to show changes in the AFP promoter of the C to A and C to T sequences previously reported (18). This observation still remains unexplained.

However, a further HP-AFP study of two additional unrelated Japanese families did exhibit the G to A substitution at nucleotide -119 in the HNF-1 binding site of the AFP promoter (20). In addition to these clinical cases, the latter investigators showed that the mutation in the AFP promoter (-119) significantly stimulated its transcriptional activity in cultured human hepatoma cells but not non-hepatoma cells and in adult mouse liver cells *in vivo*. Thus, overexpression of HNF-1 stimulated wild type and mutant AFP promoters in liver tumor, but HNF-1activation could be suppressed by nuclear factor-1 (NF-1) overexpression. It was found that the HNF-1 binding site mutation led to induction of AFP gene expression in both adult and developing liver cells, while NF-1 contributed to transcription of the AFP gene only during liver development. Since that report, methods have been reported for the rapid detection of the AFP gene promoter mutations in HP-AFP to aid in sorting out the gene mutation (19).

In summation, it has been shown that the presence of unexplained persistent elevated AFP levels in pregnancy and in normal adulthood could be suspect for the presence of HP-AFP. Even though no recent HP-AFP cases have been reported in prenatal screening programs for NTD and Trisomy-21, this trait may have long gone undetected in pregnancies experiencing "unexplained MSAFP levels" due to the lack of familial (sibling) testing. The HP-AFP condition has been reported in 19 unrelated families since the first case was described in 1983 within an NTD screening program. During that time to the present, many adult HP-AFP affected female family members of child-bearing age have been identified (21). It is thus reasonable to assume that some positive NTD screens of unexplained etiology may have gone undetected because family members of first and second degree generations had not undergone AFP serum sibling testing or AFP promoter mutation DNA sequence analysis. Analysis of AFP-levels in suspected HP-AFP could prevent unnecessary anxiety in pregnant women, faulty clinical diagnosis and inappropriate surgical treatments and decisions especially in males with reproductive and urological disorders and females with growth and puberty dysfunctions (see Houvert for Review (2)). Finally, in suspected cases of HP-AFP, clinicians should rule out the presence of hepatomas and active liver inflammation and determine liver transaminase levels.

B) <u>Assay Kit Performance:</u>

The performances of the various kits for maternal serum analytes (AFP, uE3, hCG, and Inhibin A) are presented in a bar-graph format (Figs. 7-9, 11) for each of the five MS samples. As shown in Figs. 7A and 8A, AFP and uE3 mass measurements in serum among the individual kits mostly agreed, although the values from the SIEMENS Immulite kits were about 10% lower for AFP, and 5-10% higher for uE3 than those obtained with Beckman instruments. In contrast, when the kit specific uE3 MOMs were compared, values from SIEMENS Immulite 2000/2500 and the New SIEMENS 2000/2500 ranged from 20 to 50% higher than those from Beckman (Fig. 8B). Regarding the hCG kits (Fig. 9), the two Beckman instruments (Access2 and UNICEL DXI) yielded similar mean hCG values, while the SIEMENS Immulite 2000 results were 10-20% lower than those from the other assay platforms. Finally, the method comparison for Inhibin-A displayed in Fig. 11A shows that the results from the Beckman Access/2 or Unicel were similar and that the Diagnostic Systems Lab (DSL) assay platform was 20-30% lower, and this was also true for the MS Inhibin MOM values (Fig. 11B).

Interestingly, when the AFP measurements in amniotic fluid were compared, the differences among the various methods seemed somewhat larger than in serum (Fig. 7B). In particular, results from the Abbott Asxym were 15-30% higher than from the Beckman Unicel DXI instrument were about 5-10% lower, with the results from the other instruments somewhere in between. Since these specimens are derived from actual AF samples, these levels would be comparable to real patient testing**.**

C) <u>Second Trimester Screening Software Utilized:</u>

The alpha and Benetech software packages were each used by 30% and 26%, of the labs, respectively; Robert Maciel (RMA) software was employed by 30%; and in-house and "other" softwares comprised 15%. Labs using programs classified as "other" are presumably proprietary software packages.

D) <u>First Trimester Screen:</u>

Five first trimester maternal serum mock samples are provided in the present mailout. All laboratories that are **validation-approved** and presently perform first trimester Down syndrome screening are **REQUIRED** to test

and report screen results; however, the laboratory results will **not** be graded at this time. Those laboratories not presently offering the test, nor planning to implement the test, can request that no further samples be sent to them. The FT sample (FT = first trimester) information provided to participating labs included maternal age, nuchal translucency (NT measurements in millimeters), last menstrual period (LMP), crown-rump length (CRL) measurements, race, maternal body weight, and draw date.

As demonstrated in FT Table 2, Section II, the all lab measurement of the 11.5 week Hispanic **FT271** specimen for total hCG resulted in a mass mean of 70.09 IU/ml ± 9.08, with a MOM of 0.95 (Table 2). Furthermore, the all-lab mass mean for PAPP-A was 1629.28 ± 312.86 ng/ml with a MOM of 3.24 ± 1.60. This resulted in an all-lab T21 risk assessment of 1 in 19,000 for the FT271 specimen consistent with a negative screen (Fig. 13). Thus, the FT271 sample resulted in a 100% T21 negative screen assessment.

The all lab measurement of the 11.9 week White **FT272** specimen for total hCG resulted in a mass mean of 148.04 ± 27.36 IU/ml, with a MOM of 1.99; the all-lab mass mean for PAPP-A was 798.22 ± 103.63 ng/ml with a MOM of 1.36 ± 0.68; and the all-lab T21 risk assessment was 1 in 38. The **FT272** sample resulted in a 93% T21 positive screen assessment. Further action was indicated which included genetic counseling, 93%; ultrasound, 33%; amniocentesis, 40%; and chorionic sampling, 53%. Finally, the **FT272** specimen screened negative for T18 (1 in 3,510) using a cutoff of 1 in 100 (Figs. 13, 14).

In the **FT273** Asian specimen, the gestational age all-lab mean was reported as 11.2 weeks. Assay measurements for **FT273** resulted in an all-lab total hCG mass measurement of 69.56 ± 9.59 IU/ml (MOM = 0.72 ± 0.12), while the all-lab PAPP-A mass assessment was 1407.35 ± 171.21 ng/ml (MOM = 2.05 ± 1.04). All labs agreed that the **FT273** sample was screen negative for T21 with a risk assessment of 1 in 23,900 (Fig. 13). The all-lab T18 risk assessment for **FT273** was 1 in 119,000; hence, the **FT274** specimen resulted in a negative screen for T18 (Fig. 14).

As shown in Table 2 for the **FT274** Hispanic specimen, the gestational age all-lab mean was reported as 12.4 weeks. Assay measurements resulted in an all-lab total hCG mass measurement of 59.49 ± 6.88 IU/ml (MOM = 0.83 ± 0.08) and an all-lab PAPP-A mass measurement of 2055.52 ± 412.36 ng/ml (MOM = 2.51 ± 1.22). The all-lab T21 screen consensus for **FT274** was negative with a risk assessment of 1 in 21,000. No further actions were recommended by the labs. Finally, the **FT274** specimen screened negative for T18 (1 in 119,000) using a risk cutoff of 1 in 100.

For the Hispanic **FT275** specimen, the gestational age all-lab mean was reported as 13.1 weeks. Assay measurements resulted in an all-lab total hCG mass measurement of 56.62 ± 5.63 IU/ml (MOM = 0.82 ± 0.10) while the all-lab PAPP-A mass assessment was 2307.52 ± 346.91 ng/ml (MOM = 2.01 ± 1.03). The all-lab FT T21 risk assessment was 1 in 24,000 and all labs agreed that the **FT275** sample was negative for T21 (Fig. 13). The **FT275** specimen also resulted in a negative screen for T18 with an all-lab risk assessment of 1 in 119,000.

D. 1.) <u>First Trimester Assay kit Performance:</u>

In order to compare the new Beckman Access 2/Unicel assays (60% users) for PAPP-A with those of the older Siemens Immulite and DSL assay platforms, a conversion factor was calculated from participating labs for the last five PT mailouts. Since there was a shift in the Siemens's slope from the pre-2011 PT, we used only the last 15 data points (FT samples 261-275) to calculate the conversion factor. Hence Beckman Access 2/Unicel (y-axis) data for PAPP-A ug/ml were plotted (Fig. 15A) versus Siemens Immulite 2000 (x-axis) data mIU/ml, yielding a linear correlation with an R^2 value of 0.9824 and a slope of 0.1498. In Fig. 15B, Beckmann Access2/Unicel PAPP-A values (y-axis) were plotted against DSL PAPP-A values (x-axis), yielding a second degree polynomial correlation with an R^2 value of 0.9927. Using the respective correlation equation allowed us to convert mIU/ml values into ng/ml and to directly compare Beckman Access 2/Unicel PAPP-A mass units of ng/ml to the mIU/mL mass units generated by Siemens Immulite and DSL (Fig. 12A). However, for grading purposes, each lab's results were compared to their own peer group without conversion.

Fig. 15A

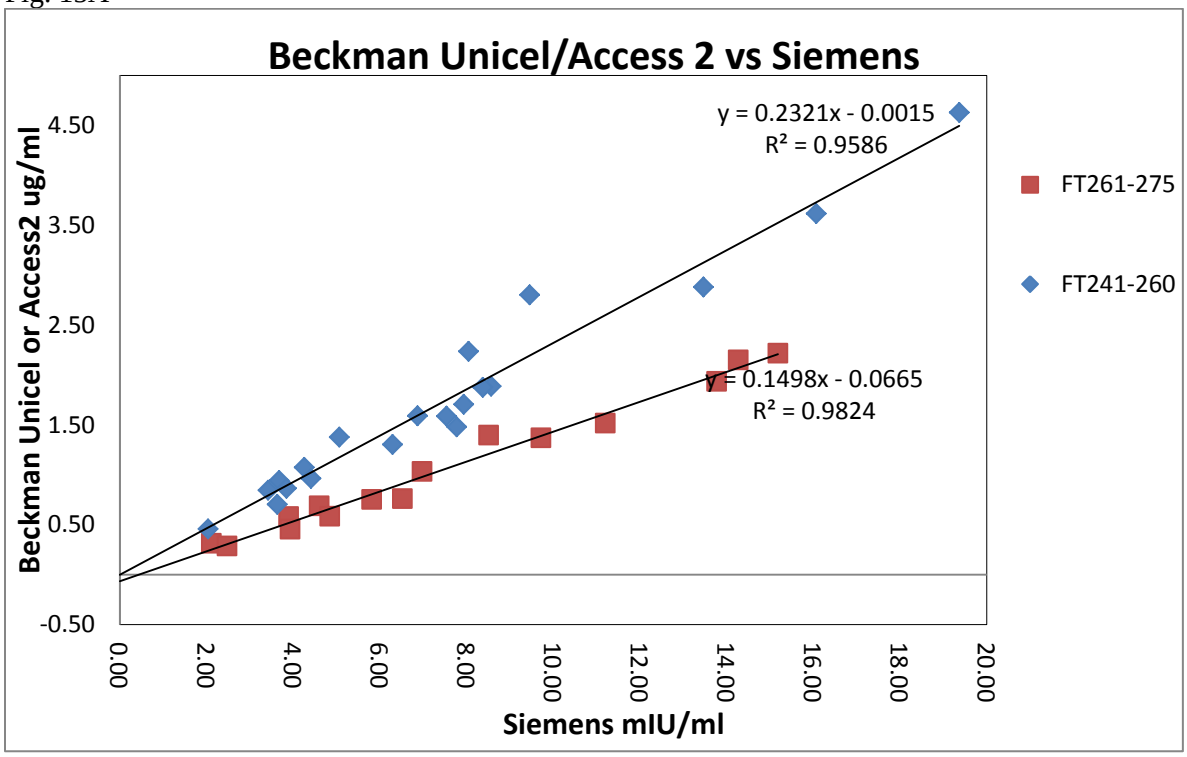
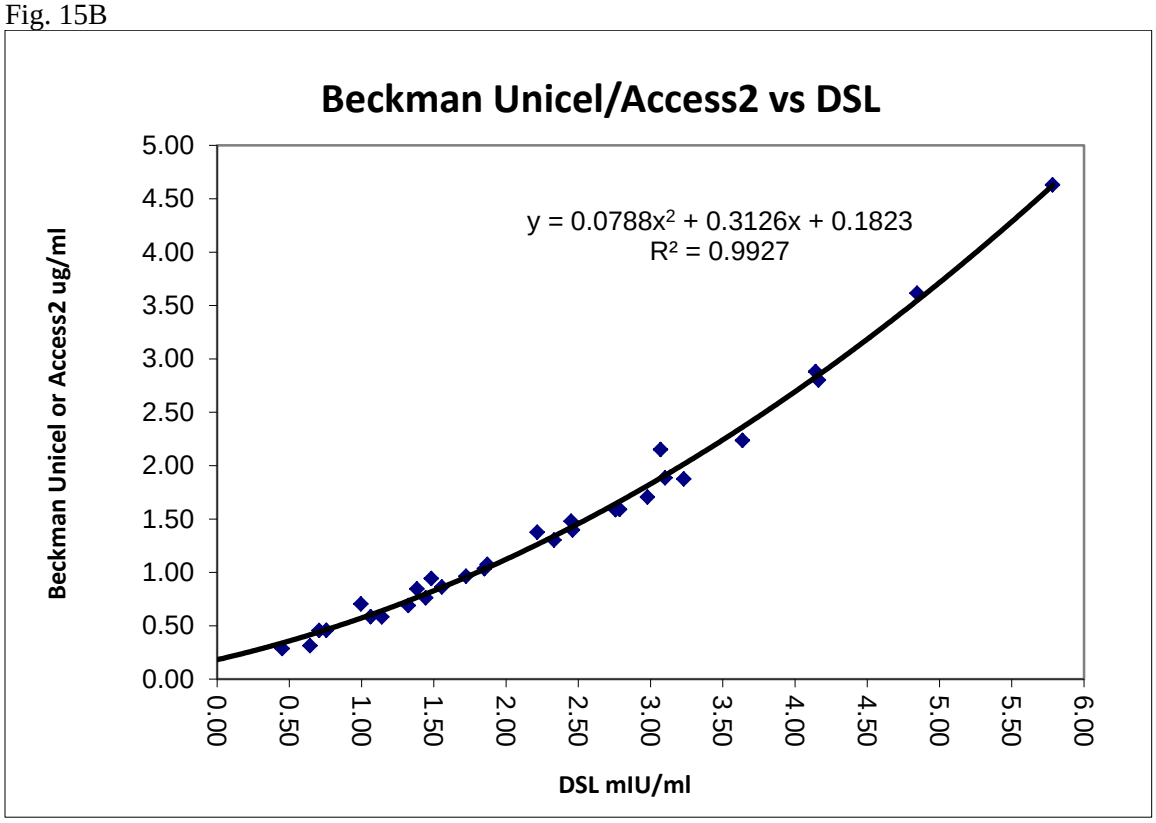


Fig. 15B



The performance of the kits used for first trimester maternal serum analytes (hCG and PAPP-A) are presented in Figs. 10 and 12 for each of the five FT samples. As shown in Fig. 10, hCG measurements between the two Beckman instruments were similar, while the Siemens Immulite instruments measured approximately 10-30% below the Beckman Access 2/Unicel instruments. The results from the three PAPP-A kits when converted to the same mass units were relatively consistent among each other with a 5% higher value for DSL. In contrast, when the PAPP-A kit MOMs were compared, Siemens Immulite were more than double those from DSL and Beckman (Fig. 12B).

E) First Trimester Screening Software Utilized:

The alpha and Benetech software packages were each used by 33% and 13% of the labs, respectively; Robert Maciel (RMA) software was employed by 33%; and in-house software comprised 20%. None of the labs used programs classified as "other" which are proprietary software packages.

G.J. Mizejewski, Ph.D.

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ABSTRACTS

A) Screening Abstract "Picks-of-the-Month":

(1) Title: The impact of severe *hyperemesis gravidarum* on the triple test screening results.

Source: J Matern Fetal Neonatal Med. 2011 Aug 8. [Epub ahead of print]

Authors: Peled Y, Melamed N, Krissi H, Eitan R, Yogev Y, Pardo J.

Abstract: Objective: we aimed to determine the influence of severe *hyperemesis gravidarum* on the interpretation of the triple test screen results.
Methods: A retrospective, case control study. The study group included 73 women who were hospitalized due to severe *hyperemesis gravidarum* and data regarding triple screening test was available. Data was compared with a control group consisting of low-risk patients without *hyperemesis gravidarum*, who underwent the triple screening test in the same laboratory and matched to the study group by maternal age and gestational age at the time of screening in a 2:1 ratio.
Results: Overall, 219 gravid patients were included in the study, of whom 73 were diagnosed with severe *hyperemesis gravidarum*. Patients in the control group were characterized by higher weight at the time of triple test screen (53.7 ± 10.9 vs. 59.7 ± 14.3 years, $p = 0.043$). No difference was found in the level of Alpha-fetoprotein or unconjugated estriol (uE3) between the groups; however the level of hCG was significantly increased in women with severe *hyperemesis gravidarum* (1.2977 ± 0.82 vs. 1.0662 ± 0.53 MoM, $p = 0.013$).
Conclusion: Increase in the level of hCG in women with severe *hyperemesis gravidarum* alter the results of triple test screen. This data should be incorporated when counseling patients regarding overall risk for chromosomal abnormalities.

(2) Title: Comparison of combined, stepwise sequential, contingent, and integrated screening in 7292 high-risk pregnant women.

Source: Prenat Diagn. 2011 Jul 29. doi: 10.1002/pd.2836. [Epub ahead of print]

Authors: Guanciali-Franchi P, Iezzi I, Palka C, Matarrelli B, Morizio E, Calabrese G, Benn P.

Abstract: Objective: To compare the efficacy of combined, stepwise sequential, and contingent screening versus the integrated test in detecting fetal aneuploidies.
Study Design: First trimester combined test, sequential second trimester, and contingent risks were retrospectively calculated for 7292 unselected pregnant women with singleton pregnancies who had received integrated screening. The first trimester testing was based on nuchal translucency, pregnancy-associated plasma protein-A, and free-beta-human chorionic gonadotrophin (free β-hCG) and the second trimester tests were alpha-fetoprotein, hCG, and unconjugated estriol. A second trimester risk of 1:250 defined a positive result for all protocols with the contingent protocol based on additional second trimester testing for those with risks between 1:30 and 1:1200.
Results: Among the population submitted for the integrated test, the detection rate was 19/21 (90%) for Down syndrome (DS) and 6/6 (100%) for Edwards syndrome (ES) and the DS false-positive rate (FPR) was 247/7271 (3.4%). Provision of the first trimester combined test alone would have resulted in a 17/21 (81%) detection rate for DS, that of 4/6 (67%) for ES and a DS FPR of 292/7271 (4.0%). The sequential and contingent approaches had the same final detection rates as the integrated test but potentially allowed a high proportion of the affected pregnancies to be detected in the first trimester. The lowest net DS FPR was seen with the contingent approach (2.6%) and using this protocol only 12.7% of women would have required second trimester testing.
Conclusions: Integrated, sequential, and contingent screenings are all more efficacious than the combined test. Overall, the contingent approach was the most efficient with a high-detection rate, the lowest FPR, and the least amount of testing.

(3) Title: Prenatal screening for fetal aneuploidy in singleton pregnancies.

Source: J Obstet Gynaecol Can. 2011 Jul;33(7):736-50.

Authors: Chitayat D, Langlois S, Wilson RD.

Abstract: Objective: To develop a Canadian consensus document on maternal screening for fetal aneuploidy (e.g., Down syndrome and trisomy 18) in singleton pregnancies.
Options: Pregnancy screening for fetal aneuploidy started in the mid 1960s, using maternal age as the screening test. New developments in maternal serum and ultrasound screening have made it possible to offer all pregnant patients a non-invasive screening test to assess their risk of having a fetus with aneuploidy to determine whether invasive prenatal diagnostic testing is necessary. This document reviews the options available for non-invasive screening and makes recommendations for Canadian patients and health care workers.
Outcomes: To offer non-invasive screening for fetal aneuploidy (trisomy 13, 18, 21) to all pregnant women. Invasive prenatal diagnosis would be offered to women who screen above a set risk cut-off level on non-invasive screening or to pregnant women whose personal, obstetrical, or family history places them at increased risk. Currently available non-invasive screening options include maternal age combined with one of the following: (1) first trimester screening (nuchal translucency, maternal age, and maternal serum biochemical markers), (2) second trimester serum screening (maternal age and maternal serum biochemical markers), or (3) 2-step integrated screening, which includes first and second trimester serum screening with or without nuchal translucency (integrated prenatal screen, serum integrated prenatal screening, contingent, and sequential). These options are reviewed, and recommendations are made.
Evidence: Studies published between 1982 and 2009 were retrieved through searches of PubMed or Medline and CINAHL and the Cochrane Library, using appropriate controlled vocabulary and key words (aneuploidy, Down syndrome, trisomy, prenatal screening, genetic health risk, genetic health surveillance, prenatal diagnosis). Results were restricted to systematic reviews, randomized controlled trials, and relevant observational studies. There were no language restrictions. Searches were updated on a regular basis and incorporated in the guideline to August 2010. Grey (unpublished) literature was identified through searching the websites of health technology assessment and health technology assessment-related agencies, clinical practice guideline collections, clinical trial registries, and national and international medical specialty societies. The previous Society of Obstetricians and Gynaecologists of Canada guidelines regarding prenatal screening were also reviewed in developing this clinical practice guideline.
Values: The quality of evidence was rated using the criteria described in the Report of the Canadian Task Force on Preventive Health Care.
Benefits, Harms, and Costs: This guideline is intended to reduce the number of prenatal invasive procedures done when maternal age is the only indication. This will have the benefit of reducing the numbers of normal pregnancies lost because of complications of invasive procedures. Any screening test has an inherent false-positive rate, which may result in undue anxiety. It is not possible at this time to undertake a detailed cost-benefit analysis of the implementation of this guideline, since this would require health surveillance and research and health resources not presently available; however, these factors need to be evaluated in a prospective approach by provincial and territorial initiatives.
Recommendations: 1. All pregnant women in Canada, regardless of age, should be offered, through an informed counselling process, the option of a prenatal screening test for the most common clinically significant fetal aneuploidies in addition to a second trimester ultrasound for dating, assessment of fetal anatomy, and detection of multiples. (I-A) 2. Counselling must be non-directive and must respect a woman's right to accept or decline any or all of the testing or options offered at any point in the process. (III-A) 3. Maternal age alone is a poor minimum standard for prenatal screening for aneuploidy, and it should not be used a basis for recommending invasive testing when non-invasive prenatal screening for aneuploidy is available. (II-2A) 4. Invasive prenatal diagnosis for cytogenetic analysis should not be performed without multiple marker screening results except for women who are at increased risk of fetal aneuploidy (a) because of ultrasound findings, (b) because the pregnancy was conceived by in vitro fertilization with intracytoplasmic sperm injection, or (c) because the woman or her partner has a history of a previous child or fetus with a chromosomal abnormality or is a carrier of a chromosome

rearrangement that increases the risk of having a fetus with a chromosomal abnormality. (II-2E) 5. At minimum, any prenatal screen offered to Canadian women who present for care in the first trimester should have a detection rate of 75% with no more than a 3% false-positive rate. The performance of the screen should be substantiated by annual audit. (III-B) 6. The minimum standard for women presenting in the second trimester should be a screen that has a detection rate of 75% with no more than a 5% false-positive rate. The performance of the screen should be substantiated by annual audit. (III-B) 7. First trimester nuchal translucency should be interpreted for risk assessment only when measured by sonographers or sonologists trained and accredited for this service and when there is ongoing quality assurance (II-2A), and it should not be offered as a screen without biochemical markers in singleton pregnancies. (I-E) 8. Evaluation of the fetal nasal bone in the first trimester should not be incorporated as a screen unless it is performed by sonographers or sonologists trained and accredited for this service and there is ongoing quality assurance. (II-2E) 9. For women who undertake first trimester screening, second trimester serum alpha fetoprotein screening and/or ultrasound examination is recommended to screen for open neural tube defects. (II-1A) 10. Timely referral and access is critical for women and should be facilitated to ensure women are able to undergo the type of screening test they have chosen as first trimester screening. The first steps of integrated screening (with or without nuchal translucency), contingent, or sequential screening are performed in an early and relatively narrow time window. (II-1A) 11. Ultrasound dating should be performed if menstrual or conception dating is unreliable. For any abnormal serum screen calculated on the basis of menstrual dating, an ultrasound should be done to confirm gestational age. (II-1A) 12. The presence or absence of soft markers or anomalies in the 18- to 20-week ultrasound can be used to modify the a priori risk of aneuploidy established by age or prior screening. (II-2B) 13. Information such as gestational dating, maternal weight, ethnicity, insulin-dependent diabetes mellitus, and use of assisted reproduction technologies should be provided to the laboratory to improve accuracy of testing. (II-2A) 14. Health care providers should be aware of the screening modalities available in their province or territory. (III-B) 15. A reliable system needs to be in place ensuring timely reporting of results. (III-C) 16. Screening programs should be implemented with resources that support audited screening and diagnostic laboratory services, ultrasound, genetic counseling services, patient and health care provider education, and high quality diagnostic testing, as well as resources for administration, annual clinical audit, and data management. In addition, there must be the flexibility and funding to adjust the program to new technology and protocols. (II-3B).

(4) Title: Impact of smoking on maternal serum markers and prenatal screening in the first and second trimesters.

Source: Prenat Diagn. 2011 Jun;31(6):583-8. doi: 10.1002/pd.2755. Epub 2011 Apr 11.

Authors: Zhang J, Lambert-Messerlian G, Palomaki GE, Canick JA.

Abstract: Objectives: To examine the effects of smoking on first and second trimester screening markers and to determine the overall impact of these effects on Down syndrome and trisomy 18 risks in first trimester combined, second trimester quadruple and integrated tests.
Methods: Examination of screening records at Women and Infants Hospital during 2006-2008. First trimester pregnancy-associated plasma protein-A (PAPP-A), beta-human chorionic gonadotrophin (hCG) and nuchal translucency and second trimester alpha-fetoprotein (AFP), unconjugated estriol (uE3), hCG and inhibin A (inhA) multiple of the median (MoM) values were extracted from the database along with risk results, smoking status and relevant demographic information.
Results: Smoking led to significantly reduced median levels of first trimester PAPP-A (0.89 MoM) and hCG (0.80 MoM), reduced second trimester uE3 (0.96 MoM) and hCG (0.84 MoM), and increased AFP (1.03 MoM) and inhA (1.39 MoM). After accounting for the differences in age between groups, smokers had higher Down syndrome screen positive rates for the second trimester quadruple test, but not for first trimester combined or integrated tests. Screen positive rates for trisomy 18 were markedly increased in smokers relative to age-matched non-smokers when using first trimester combined or integrated tests.
Conclusion: Smoking leads to increased screen positive rates, especially for trisomy 18 using combined or integrated tests.

B) Case History Screening "Picks-of-the-Month":

(1) Title: Prenatal diagnosis and molecular cytogenetic characterization of a derivative chromosome der(18;18)(q10;q10)del(18)(q11.1q12.1)del(18)(q22.1q22.3) presenting as apparent isochromosome 18q in a fetus with holoprosencephaly.

Source: Taiwan J Obstet Gynecol. 2011 Jun;50(2):182-187.

Authors: Chen CP, Kuo YK, Su YN, Chern SR, Tsai FJ, Wu PC, Chen YT, Town DD, Wang W.

Abstract: Objective: To present prenatal diagnosis and molecular cytogenetic characterization of a derivative chromosome der(18;18)(q10;q10)del(18)(q11.1q12.1)del(18)(q22.1q22.3).
Materials, Methods and Results: A 32-year-old woman was referred for genetic counseling of prenatally detected isochromosome 18q [i(18q)]. She had undergone amniocentesis at 19 gestational weeks because of a trisomy 18 risk of 1/39 derived from abnormally low levels of maternal serum unconjugated estriol, inhibin A, α-fetoprotein, and total β-human chorionic gonadotropin. Amniocentesis revealed a karyotype of 46,XX,i(18)(q10). Parental karyotypes were normal. Prenatal ultrasound showed alobar holoprosencephaly. Repeated amniocentesis was requested and performed at 21 gestational weeks. Array-comparative genomic hybridization analyses revealed a 14-Mb deletion of 18p11.32-p11.21, a 37.8-Mb duplication of 18q12.1-q22.1, and a 6.9-Mb duplication of 18q22.3-q23. Metaphase fluorescence in situ hybridization study showed the absence of an 18q12.1-specific probe signal in one arm and the absence of an 18q22.2-specific probe signal in the other arm of the derivative chromosome. Quantitative fluorescent polymerase chain reaction analysis determined a paternal origin of the derivative chromosome. The cytogenetic result was 46,XX,der(18;18)(q10;q10)del(18)(q11.1q12.1)del(18)(q22.1q22.3). The fetus postnatally manifested cebocephaly.
Conclusion: Concomitant monosomy 18p and trisomy 18q can be associated with holoprosencephaly and abnormal maternal serum screening results. Array-comparative genomic hybridization, fluorescence in situ hybridization, and quantitative fluorescent polymerase chain reaction are useful in genetic counseling of prenatally detected isochromosomes by providing information on the origin and genetic components of the isochromosome.

(2) Title: Placental mesenchymal dysplasia, a case of intrauterine sudden death of fetus with rupture of cirsoid periumbilical chorionic vessels.

Source: Diagn Pathol. 2011 Apr 24;6:38..

Authors: Umazume T, Kataoka S, Kamamuta K, Tanuma F, Sumie A, Shirogane T, Kudou T, Ikeda H.

Abstract: We report a 32-year-old woman (1-gravid, 1-para) with a vesicular lesion in her uterus that was pointed out on ultrasound at 8 weeks' gestation. Amniocentesis at 15 weeks' gestation showed a normal female karyotype, 46XX. As the pregnancy advanced, the mole-like lesion became relatively reduced. Throughout gestation, the maternal human chorionic gonadotropin level was normal, but the serum alpha fetoprotein level rose as her pregnancy progressed. Her fetus did not exhibit any remarkable anomalies. The patient visited our hospital complaining of a diminished feeling of fetal movements at 36 weeks 5 days' gestation, and intrauterine fetal death (IUFD) was confirmed. She delivered a 2336-g female without any definite anomalies. A pathological examination led to a diagnosis of placental mesenchymal dysplasia, and androgenetic/biparental mosaicism in the placenta was identified using p57kip2 immunohistochemical staining. And it also revealed that the rupture of the cirsoid chorionic vessels had led to IUFD.

(3) Title: Placental characteristics as a proxy measure of serum hormone and protein levels during pregnancy with a male fetus.

Source: Cancer Causes Control. 2011 May;22(5):689-95. Epub 2011 Feb 19.

Authors: Trabert B, Longnecker MP, Graubard BI, Klebanoff MA, Stanczyk FZ, McGlynn KA.

Abstract: Objective: In utero exposure to steroid hormones may be related to risk of some cancers such as testicular germ cell tumors (TGCT). To determine whether placental characteristics are good surrogate measures of maternal biomarker levels, we evaluated the correlations in mothers of sons at higher (whites, n = 150) and lower (blacks, n = 150) risk of TGCT. Associations with birth weight were also examined.
Methods: All mothers, participants in the Collaborative Perinatal Project, were primigravidas who gave birth to male singletons. Associations between placental weight and placental thickness and third-trimester biomarker levels were evaluated using linear regression. Partial correlation coefficients for placental characteristics and birth weight were also estimated.
Results: Placental weight was positively correlated with alpha-fetoprotein (AFP), sex hormone-binding globulin (SHBG), testosterone, estradiol and estriol in whites, and AFP and estriol in blacks. Placental thickness was not associated with any biomarker. After adjustment for placental weight, birth weight was not correlated with any biomarker.
Conclusions: In these data, placental weight was modestly correlated with third-trimester biomarker level; however, it appeared to be a better surrogate for third-trimester biomarker level than birth weight. Placental thickness had limited utility as a surrogate measure for biomarker levels.

(4) Title: Reasons for adult referrals for genetic counseling at a genetics center in Izmir, Turkey: analysis of 8965 cases over an eleven-year period.

Source: J Genet Couns. 2011 Jun;20(3):287-93. doi: 10.1007/s10897-010-9342-9. Epub 2011 Jan 8.

Authors: Cogulu O, Ozkinay F, Akin H, Onay H, Karaca E, Durmaz AA, Durmaz B, Aykut A, Pariltay E, Kirbiyik O, Gunduz C, Ozkinay C.

Abstract: A limited numbers of published studies evaluate the referral reasons for genetic counseling services in the literature. These studies are focused on prenatal genetic counseling services, in particular, prenatal diagnosis. In order to provide the most effective and helpful genetic counseling services, genetics professionals need adequate knowledge about the profile of individuals referred for these services. In addition, physicians need increased awareness of the nature of genetic issues in order to make appropriate referrals. This study was intended to provide a descriptive analysis of the referral reasons of patients that received genetic counseling at a genetics center in Izmir, Turkey during an 11-year period. A total of 8965 records generated between 1998 and 2008 from one genetic center (which consists of The Department of Medical Genetics and Division of Pediatric Genetics) were evaluated retrospectively. Of these, 6,258 involved referrals for prenatal reasons, and 2,707 involved referrals for postnatal reasons. Both prenatal and postnatal records were further classified into more specific categories of referral reasons. The most common reason for genetic counseling among the prenatal patients was advanced maternal age (42.0%), followed by high risk results on prenatal biochemical screening tests such as second trimester double test [(serum concentration of alphafetoprotein (AFP), beta-human chorionic gonadotropin (beta-HCG)], triple test (serum concentration of AFP, beta-HCG, oestriol) and integrated test (26.5%). The most common indications for postnatal patients were recurrent miscarriages (28.2%) and infertility (19.7%). A significant increase in number of specific categories of referrals for genetic counseling was observed for the last 3 years after the establishment of the Medical Genetics Department. These data provide useful information about the frequency of referrals to the genetics department, and the feasibility of genetic services. Organization of genetic services and systematic procedures for genetic counseling and genetic testing may improve the public's awareness of genetics and ensure a high standard of patient care.

C) News of Note: Abstract of New Markers:

(1) Title: Second Trimester Prenatal Screening for Down's Syndrome in Mainland Chinese Subjects using Double-Marker Analysis of α-fetoprotein and β-human Chorionic Gonadotropin Combined with Measurement of Nuchal Fold Thickness.

Source: Ann Acad Med Singapore. 2011 Jul;40(7):315-4.

Authors: Liu F, Liang H, Jiang X, Zhang Y, Xue L, Yang C, Cheng J, Liu P, Liu Y, Guo X.

Abstract: Introduction: This study examines the effectiveness of double-marker analysis for α-fetoprotein (AFP) and β-human chorionic gonadotropin (β-hCG) combined with measurement of nuchal fold thickness (NT) in the detection of Down's syndrome (DS) in Mainland Chinese subjects during second trimester prenatal screening.
Materials and Methods: We examined pregnant women with a singleton pregnancy between 15 and 21 weeks of gestation who underwent second trimester screening for DS using double-marker analysis for AFP and β-hCG combined with ultrasound measurement of NT. The combined risk of DS was calculated. A cut-off of 1/270 was used to defi ne a pregnancy at high-risk of DS. Amniocentesis was offered to all patients with high-risk pregnancies.
Results: Using double-marker analysis for AFP and β-hCG in combination with measurement of NT, the detection rate of DS increased from 66.7% to 77.8% when compared with double-marker analysis alone with similar false-positive rates (4.35%, 4.83% respectively). Using receiver operating characteristic curve (ROC) analysis, we determined that the double-marker analysis combined with measurement of NT exhibited an increased area under the curve (AUC) of 0.835 (95% CI: 0.743 to 0.927) when compared to double-marker analysis alone, which had an AUC of 0.748 (95% CI: 0.635 to 0.860). In addition, both methods were more effective than any other single test such as AFP, free β-hCG or NT measurement.
Conclusion: Second trimester prenatal screening using double-marker analysis for AFP and β-hCG combined with measurement of NT is effective for the detection of DS in Mainland Chinese pregnancies.

(2) Title: The association between second-trimester maternal serum alpha-fetoprotein in 14-22 weeks and adverse pregnancy outcome.

Source: Acta Med Iran. 2010 Jul-Aug;48(4):234-8.

Authors: Dehghani-Firouzabadi R, Tayebi N, Ghasemi N, Tahmasbi Z.

Abstract: Aim of this study is to determine the risk of adverse pregnancy outcome by maternal serum alpha-fetoprotein (MSAFP) level. We followed 295 pregnant women from MSAFP screening in the 14th to 22th week of gestation until the end of pregnancy and information on pregnancy outcome have been recorded in questionnaires. Of 295 pregnant women, 270 had term labor and 25 had preterm labor. The frequencies of pregnancy outcomes were as following: 3 (1.01%) stillbirths, 25 (8.47%) preterm labor, and 10 (3.4%) preterm rupture of membranous (PROM), 15 (5.1%) pre-eclampsia, 23 (7.8%) oligohydramnious, and 1 (0.33%) miscarriage. The mean of preterm labor was significantly associated with the higher level of MSAFP ($P = 0.021$). The mean was 55.1 ng/cc in preterm labor and 41.1 ng/cc in term labor. Also, second trimester MSAFP levels were higher in women with pre-eclampsia ($P < 0.001$). The significant association was found between higher level of MSAFP with oligohydramnious ($P < 0.001$) and low birth weight ($P < 0.001$). Pregnancies with an elevated MSAFP level are associated with adverse obstetric outcomes and need more prenatal care.

(3) Title: Use of ethnic-specific medians for Hispanic patients reduces ethnic disparities in multiple marker screening.

Source: Prenat Diagn. 2011 Apr;31(4):331-3. doi: 10.1002/pd.2650. Epub 2011 Jan 20.

Authors: Wetta L, Biggio J Jr, Owen J.

Abstract: Objective: To estimate whether midtrimester maternal serum analyte concentrations differ between Caucasian and Hispanic women and whether using ethnic-specific medians affects quad screen performance.
Method: Caucasian and Hispanic patients with singletons who underwent maternal serum screening in our laboratory were identified. Alfa-fetoprotein (AFP), estriol, human chorionic gonadotrophin (hCG), and inhibin-A medians were derived separately for Caucasians, Hispanics, and for the composite group. Using composite medians, intergroup mean multiples of the medians (MoMs) for each analyte were compared. Using ethnic-specific medians, new MoMs were calculated and utilized in a risk estimation algorithm.
Results: A total of 5478 Caucasian and 2246 Hispanic pregnancies were evaluated. Intergroup MoMs were significantly different for all analytes. AFP, hCG, and inhibin-A were lower in Hispanics, while estriol was higher ($P < 0.0001$). Using composite medians, the screen-positive rate (SPR) for trisomy 21 was 5.39% in Caucasians and 3.29% in Hispanics. Ethnic-specific medians reduced this disparity: 4.76% in Caucasians and 4.05% in Hispanics. The SPR for neural tube defects with composite medians was 1.44% for Caucasians and 0.89% for Hispanics; with ethnic-specific medians, the SPR was 1.42% for Caucasians and 1.07% for Hispanics.
Conclusions: Serum analyte concentrations differ between Caucasian and Hispanic gravidas. Use of ethnic-specific medians reduces the disparity in SPR for trisomy 21 and neural tube defects.

(4) Title: The Reliability of Maternal Serum Triple Test in Prenatal Diagnosis of Fetal Chromosomal Abnormalities of Pregnant Turkish Women.

Source: Genet Test Mol Biomarkers. 2011 Jun 23. [Epub ahead of print]

Authors: Demirhan O, Pazarbaşı A, Güzel AI, Taştemir D, Yılmaz B, Kasap M, Ozgünen FT, Evrüke C, Demir C, Tunç E, Kocatürk-Sel S, Onatoğlu-Arıkan D, Koç S, Ozer O, Inandıklıoğlu N.

Abstract: Aim: The purpose of this article was to evaluate the reliability of maternal serum triple marker screening of alpha-fetoprotein, human chorionic gonadotropin, and unconjugated estriol for the prenatal diagnosis of fetal chromosomal abnormalities in Turkish pregnant women.
Method: Medical records were used to analyze indications of amniocentesis and quantitative fluorescent-polymerase chain reaction. Anomaly screening was performed for all patients between 13 and 22 weeks of pregnancy. A total of 1725 pregnancies with chromosomal abnormality risk according to triple test screening were accepted for fetal chromosome analysis and quantitative fluorescent-polymerase chain reaction.
Results: Chromosomal aberrations were observed in 56 (3.2%) cases. About 44.6% of the abnormalities detected were numerical aberrations; however, 55.3% of the abnormalities were structural aberrations. Abnormalities detected were inversion of chromosome 9 in 20 cases, trisomy 21 in 14 cases, 46,XX/47,XX, +21 in 1 case, trisomy 18 in 2 cases, trisomy 13 in 1 case, 47,XXY, in 1 case, 45,X, in 1 case, structural abnormalities in 12 cases, and mosaic or tetraploidy in 6 cases.
Conclusion: Second trimester triple test is an effective screening tool for detecting fetal Down syndrome in Turkish women.

D) News of Note: Abstracts of New Testing Agents/Methods:

(1) Title: GoldMag nanocomposite-functionalized graphene sensing platform for one-step electrochemical immunoassay of alpha-fetoprotein.

Source: Biosens Bioelectron. 2011 Oct 15;28(1):174-80. Epub 2011 Jul 19.

Authors: Zhang B, Tang D, Liu B, Chen H, Cui Y, Chen G.

Abstract: A new flow-through electrochemical immunosensor was designed for sensitive detection of alpha-fetoprotein (AFP) in human serum by using nanogold-functionalized magnetic graphene nanosheets as immunosensing probes. Initially, amino functionalized magnetic beads were covalently immobilized on

the surface of graphene oxide nanosheets (MGPs), then nanogold particles were adsorbed on the amino groups of the MGPs to construct GoldMag nanocomposites functionalized graphene nanosheets (GMGPs), and then horseradish peroxidase-anti-AFP conjugates (HRP-anti-AFP) were assembled onto the surface of nanogold particles (bio-GMGP). With the aid of an external magnet, the formed bio-GMGPs were attached onto the base electrode in the flow system. With a non-competitive immunoassay format, the injected sample containing AFP antigens was produced transparent immunoaffinity reaction with the immobilized HRP-anti-AFP on the bio-GMGPs. The formed immunocomplex inhibited partly the active center of HRP, and decreased the labeled HRP toward the reduction of H(2)O(2). The performance and factors influencing the performance of the immunosensor were investigated in detail. Under optimal conditions, the electrochemical immunosensor displayed a wide working range of 0.01-200ngmL(-1) with a low detection limit (LOD) of 1.0pgmL(-1) AFP (at 3s(B)). Intra- and inter-assay coefficients of variation (CV) were below 10%. In addition, the methodology was validated with real serum samples, receiving a good correlation with the results obtained from commercially available electrochemiluminescence automated analyzer.

(2) Title: Sensitive sandwich electrochemical immunosensor for alpha fetoprotein based on prussian blue modified hydroxyapatite.

Source: Biosens Bioelectron. 2011 Oct 15;28(1):112-6. Epub 2011 Jul 18.

Authors: Dai Y, Cai Y, Zhao Y, Wu D, Liu B, Li R, Yang M, Wei Q, Du B, Li H.

Abstract: A sandwich electrochemical immunosensor for the sensitive determination of alpha fetoprotein (AFP) has been fabricated. Prussian blue modified hydroxyapatite (PB@HAP) was firstly prepared and used as electrochemical label due to the wonderful conductivity and good biocompatibility of HAP. The results proved that the immunosensor fabricated using the label based on PB@HAP loaded with horse radish peroxidase (HRP) and secondary anti-AFP antibody (Ab(2)) (PB@HAP-HRP-Ab(2)) had high sensitivity, and the sensitivity of the label PB@HAP-HRP-Ab(2) was much higher than labels of PB@HAP-Ab(2), PB-HRP-Ab(2) and HAP-HRP-Ab(2). The mixture of graphene sheet (GS) and thionine (TH) was not only used to immobilize anti-AFP antibody (Ab(1)) but also took part in the signal amplification. The amperometric signal increased linearly with AFP concentration in the range of 0.02-8ng/mL with a low detection limit of 9pg/mL. The immunosensor had the advantages of high sensitivity, good selectivity and good stability, and was applied to the analysis of AFP in serum sample with satisfactory results. Due to the low-cost and easy synthesis of PB@HAP, the screen-printed electrodes could be used instead of the bare glass carbon electrode in order to achieve mass production. In addition, it had potential application in the detection of other tumor markers.

(3) Title: A label-free immunosensor based on modified mesoporous silica for simultaneous determination of tumor markers.

Source: Biosens Bioelectron. 2011 Jul 30. [Epub ahead of print]

Authors: Lin J, Wei Z, Mao C.

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Abstract: A label-free multiplexed immunoassay strategy was proposed for the simultaneous detection of two tumor markers, carcinoembryonic antigen (CEA) and α-fetoprotein (AFP). Monoclonal antibody of CEA was co-immobilized with ferrocenecarboxylic acid (FCA) inside the channels of mesoporous silica (MPS) to prepare the label-free probe for CEA. Also, monoclonal antibody of AFP was co-immobilized with horseradish peroxidase (HRP) inside the channels of MPS to prepare the label-free probe for AFP by using o-phenylenediamine (OPD) and H(2)O(2) as the electrochemical substrates. Thus, the multianalyte immunosensor was constructed by coating the probes of CEA and AFP respectively onto the different areas of indium-tin oxide (ITO) electrode. When the immunosensor was incubated with sample antigens, CEA and AFP antigens were introduced into the mesopores of MPS after the immunoassay reaction. Because all of the Si-OH groups on the external surface of MPS were blocked with Si(CH(3))(3), the proteins and substrates were limited to be embedded on the internal pore walls. Therefore, the electric response transfer was confined inside the pore channels. The nonconductive

immunoconjugates blocked the electron transfer and the peak responses changed on the corresponding surface respectively. Then, the simultaneous detection of CEA and AFP achieved. The linear ranges of CEA and AFP were 0.5-45ngmL(-1) and 1-90ngmL(-1) with the detection limits of 0.2ngmL(-1) and 0.5ngmL(-1) (S/N=3), respectively. The fabricated immunosensor shows appropriate sensitivity and offers an alternative to the multianalyte detection of antigens or other bioactive molecules.

(4) Title: Immunodevice for simultaneous detection of two relevant tumor markers based on separation of different microparticles by dielectrophoresis.

Source: Biosens Bioelectron. 2011 Oct 15;28(1):443-9. Epub 2011 Aug 4.

Authors: Ramón-Azcón J, Yasukawa T, Mizutani F.

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Abstract: In this study, a rapid immunosensing system has been developed for simultaneous analysis of two tumor markers, alpha-fetoprotein (AFP) and prostate-specific antigen (PSA). The strategy for rapid multisensing is based on rapid immunoreactions occurring on the surface of microparticles and the spatial separation of different particles that exhibit distinct dielectrophoretic (DEP) properties. Recognition events for immunoreactions have been performed on the surfaces of two different microparticles conjugated with two different antibodies: polystyrene (PS) microparticles with an anti-AFP antibody and gold-coated (50nm) PS microparticles with an anti-PSA antibody. The DEP devices consisted of an upper indium tin oxide (ITO) glass and a lower ITO electrode with a castellated structure. Sandwich structured immunocomplexes of AFP and PSA were created on the microparticles and then labeled with fluorescent molecules via a secondary antibody. After introducing the particles into the DEP devices, an alternating current (AC) voltage (20V peak-to-peak voltage and 30kHz) was applied between the upper ITO and lower electrodes to manipulate the particles with negative dielectrophoresis (n-DEP).The uncoated PS particles and the gold-coated PS particles rapidly moved and separated to form wave-like line and triangular aggregates, respectively. The measurements of the fluorescence signals from the uncoated and gold-coated PS particles directed to different regions of the DEP device permit the determination of the concentrations of AFP and PSA simultaneously. No cross-reactivity was observed for either of the immunorecognition events. Limits of detection achieved for the AFP and PSA assays were 0.18 and 1.1ngmL(-1), respectively, which satisfy medical requirements for both antigens in human serum. The total assay time required for the simultaneous detection of the two different analytes in this study (25min) was shortened compared to the conventional enzyme-linked immunosorbent assay.

E) Special Abstract Selection:

(1) Title: Prenatal screening for trisomy 21: recent advances and guidelines.

Source: Clin Chem Lab Med. 2011 Jul 27. [Epub ahead of print]

Authors: Canick J.

Abstract: The performance of prenatal screening tests for the identification of trisomy 21 (Down syndrome) has markedly improved since the 1970s and early 1980s when maternal age was the sole mode of screening the general pregnant population. With the discovery of second trimester serum markers in the 1980s and 1990s and implementation of double, triple, and quad marker testing; the discovery of first trimester serum and ultrasound markers in the 1990s and implementation of the combined test; and the development of the integrated test and sequential screening strategies over the past decade, the performance of screening has improved to a detection rate of 90%-95% at a false positive rate of 2%-5%. In this review, I will describe the advances in prenatal screening for trisomy 21, present current screening strategies, and discuss guidelines published by professional societies and regulatory bodies, with a focus on current prenatal screening practice in the USA.

(2) Title: Unfractionated heparin for second trimester placental insufficiency: a pilot randomized trial.

Source: J Thromb Haemost. 2011 Aug;9(8):1483-92. doi: 10.1111/j.1538-7836.2011.04407.x.

Authors: Kingdom JC, Walker M, Proctor LK, Keating S, Shah PS, McLeod A, Keunen J, Windrim RC, Dodd JM.

Abstract: Objective: To conduct a pilot randomized controlled trial of unfractionated heparin (UFH) in women considered at high risk of placental insufficiency in the second trimester.
Methods: Women with either false-positive first trimester (pregnancy-associated placental protein-A [PAPP-A] <0.35 MoM) or second trimester (alpha-fetoprotein [AFP] > 2.0 MoM, inhibin > 3.0 MoM, human chorionic gonadotropin >4.0 MoM) serum screening tests or medical/obstetric risk factors were screened for placental insufficiency by sonographic evaluation of the placenta and uterine artery Doppler between 18 and 22 weeks. Thrombophilia screen-negative women with two or three abnormal test categories were randomized by 23+6 weeks to self-administration of subcutaneous unfractionated heparin (UFH) 7500 IU twice daily until birth or 34 weeks, or to standard care. Maternal anxiety and other maternal-infant outcomes were determined.
Results: Thirty-two out of 41 eligible women consented, with 16 women randomized to UFH and 16 to standard care. There was no statistically significant difference identified between the two treatment groups (standard care vs. UFH) for the following: maternal anxiety score (mean [standard deviation]), 14.2 [±1.6] vs. 14.0 [±1.8]; birth weight (median [range]), 1795 [470-3295]g vs. 1860 [730-3050]g; perinatal death, 3 vs. 0; severe preeclampsia, 2 vs. 6; placental weight <10th percentile, 7 vs. 4; or placental infarction, 4 vs. 3.
Conclusion: Our study design identified women at high risk of adverse maternal-infant outcomes attributable to placental insufficiency. Women with evidence of placental insufficiency were willing to undergo randomization and self-administration of UFH without increased maternal anxiety.

(3) Title: α-Fetoprotein.

Source: Arch Dis Child Educ Pract Ed. 2011 Aug;96(4):141-7. doi: 10.1136/adc.2011.213181. Epub 2011 May 25.

Authors: Murray MJ, Nicholson JC.

Abstract: α-Fetoprotein (AFP) measurements have clinical implications in fetal medicine and, in infants and older children, in detection, differential diagnosis and monitoring of malignant disease. Maternal serum AFP levels constitute part of a multiple-marker test used in early second-trimester screening to predict risk of fetal chromosomal abnormalities. Those individuals with increased risk are offered further definitive diagnostic investigation. Second-trimester screening is now increasingly being superseded by first-trimester screening with other serum markers and ultrasound. As AFP is only produced physiologically during fetal development, elevated serum levels after the first two post-natal years usually indicate the presence of a malignant disease process. Before this time, levels may be purely physiological and therefore serial values should be plotted on a logarithmic chart to ensure that they are falling appropriately, with a typical half-life of ~5-6 days. If not, further investigation should be undertaken. Serum AFP is raised in a significant proportion of germ cell tumours (GCTs), hepatoblastoma and hepatocellular carcinoma (HCC). In suspected cases of GCT, serum human choriogonadotropin (HCG) estimation should also be performed. For possible intracranial GCTs, both serum and cerebrospinal fluid levels of AFP and HCG should be measured, ideally before neurosurgical biopsy. In malignant conditions, serum AFP may be used for diagnosis, treatment monitoring, surveillance for disease recurrence and prognostication. Immunohistochemistry for AFP using antibody staining is routinely used to assist pathological diagnosis on tissue sections where the differential includes GCT, hepatoblastoma and/or HCC. Elevations of serum AFP also occur in non-malignant conditions such as chronic liver disease.

(4) Title: Maternal Serum α-Fetoprotein at 11-13 Weeks' Gestation in Spontaneous Early Preterm Delivery.

Source: Fetal Diagn Ther. 2011 Mar 11. [Epub ahead of print]

Authors: Beta J, Bredaki FE, Calvo JR, Akolekar R, Nicolaides KH.

Abstract: Objective: To examine the potential value of maternal serum level of α-fetoprotein (AFP) in the first trimester of pregnancy in the prediction of spontaneous early preterm delivery.
Methods: Maternal serum concentration of AFP at 11-13 weeks' gestation was measured in a case-control study of singleton pregnancies delivering phenotypically normal neonates, including 33 cases with spontaneous delivery before 34 weeks and 99 matched controls delivering after 37 weeks. The median multiple of the median (MoM) serum AFP in the two outcome groups was compared and the bivariate gaussian distributions were simulated in a previously described screened population of 33,370 pregnancies to estimate the performance of screening for early delivery by a combination of maternal characteristics and obstetric history with serum AFP.
Results: In the preterm delivery group compared to the term delivery group, the median serum AFP MoM was higher (1.33 vs. 0.97, p = 0.006). The estimated detection rate of preterm delivery, at a false-positive rate of 10%, from maternal characteristics and obstetric history was 27.5% and this increased to 36.0% with the addition of serum AFP.
Conclusions: Measurement of serum AFP at 11-13 weeks improves the prediction of early preterm delivery provided by maternal characteristics and obstetric history.

VI. Potentially helpful website connections/locations:

1) http://health.allrefer.com/health/alpha-fetoprotein-info.html

2) www.healthopedia.com/alpha-fetoprotein

3) http://pregnancy.about.com/cs/afp/a/afptesting.htm

4) http://www.webmd.com/baby/alpha-fetoprotein-afp-in-blood

5) http://pregnancy.about.com/od/afp/Alphafetoprotein_Testing.htm

6) http://www.americanpregnancy.org/prenataltesting/afpplus.html

Figure 1

Maternal Sera AFP MoM

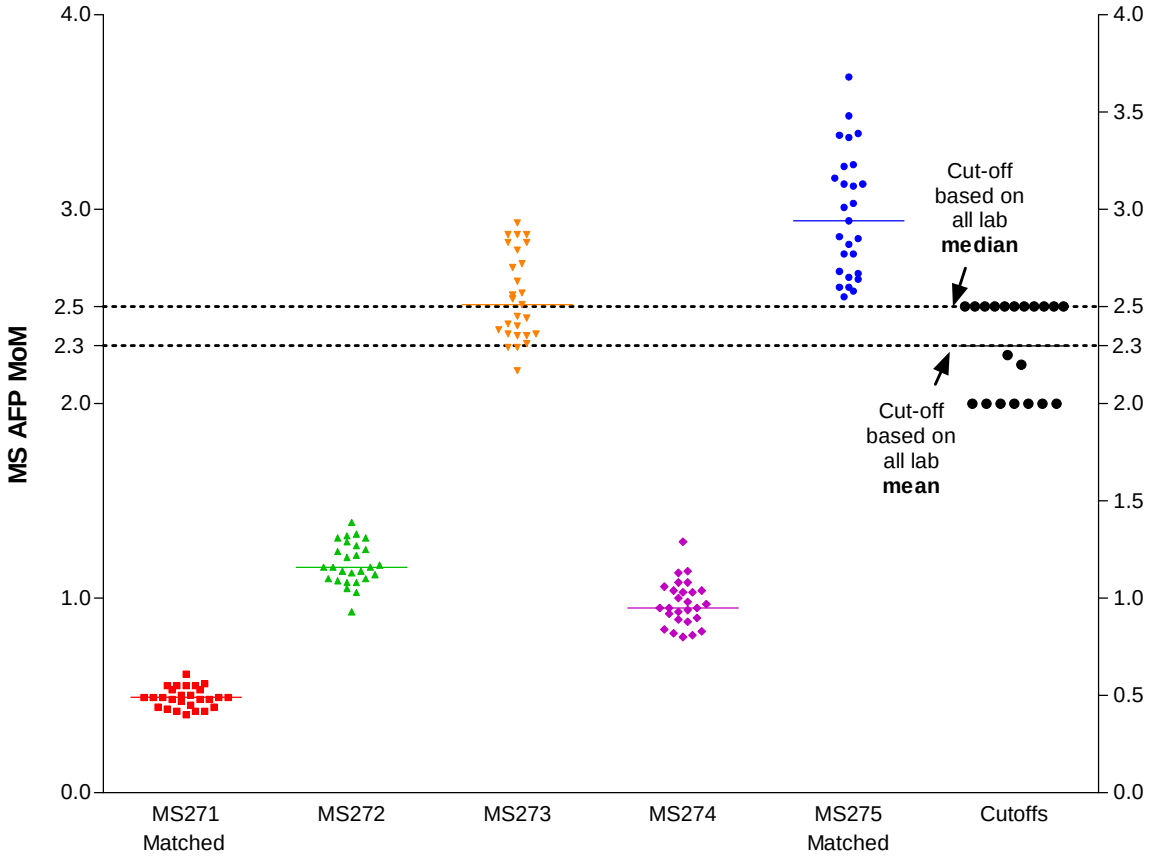


Figure 2

Amniotic Fluid AFP MoM

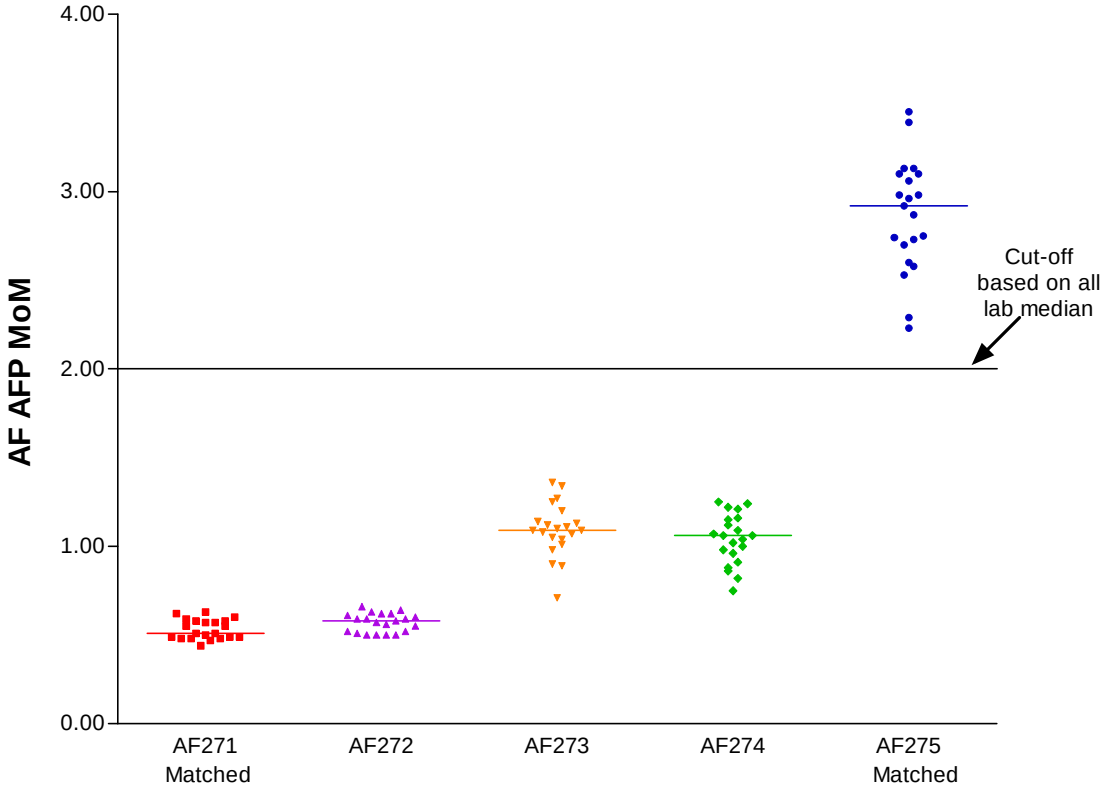


Figure 3

Graphic Distribution of Second Trimester Neural Tube Defect Risk Estimates

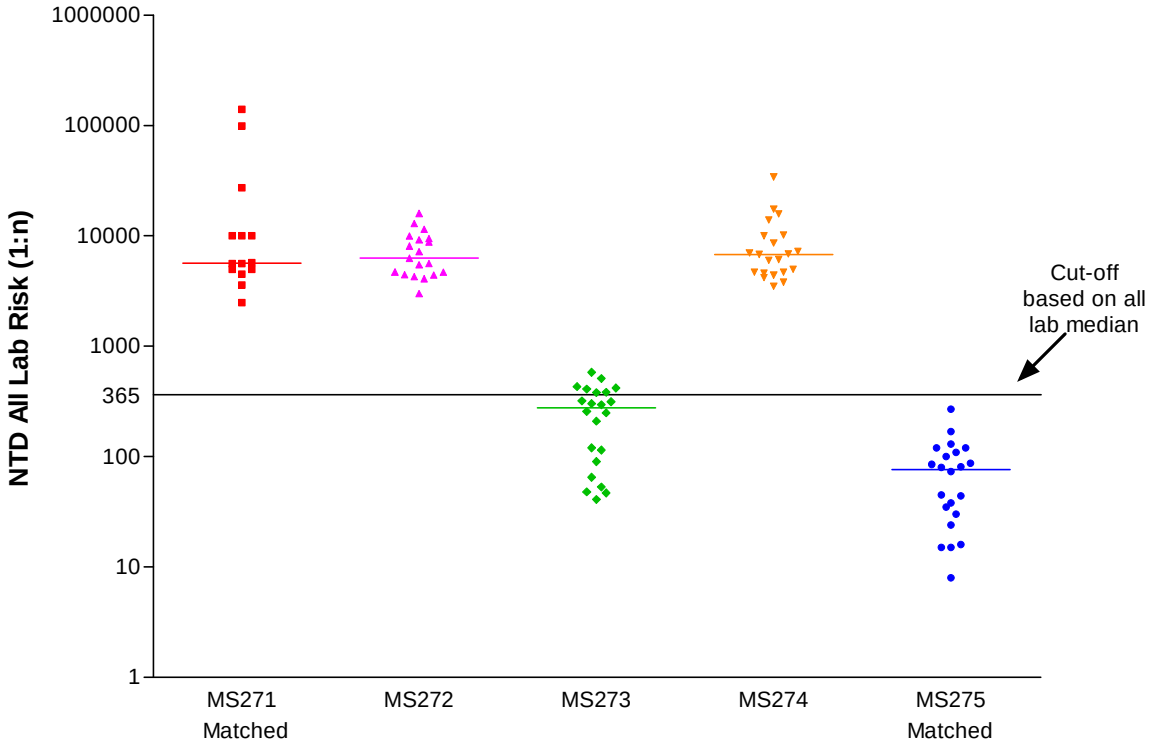


Figure 4

Graphic Distribution of Second Trimester Trisomy 18 Risk Estimates

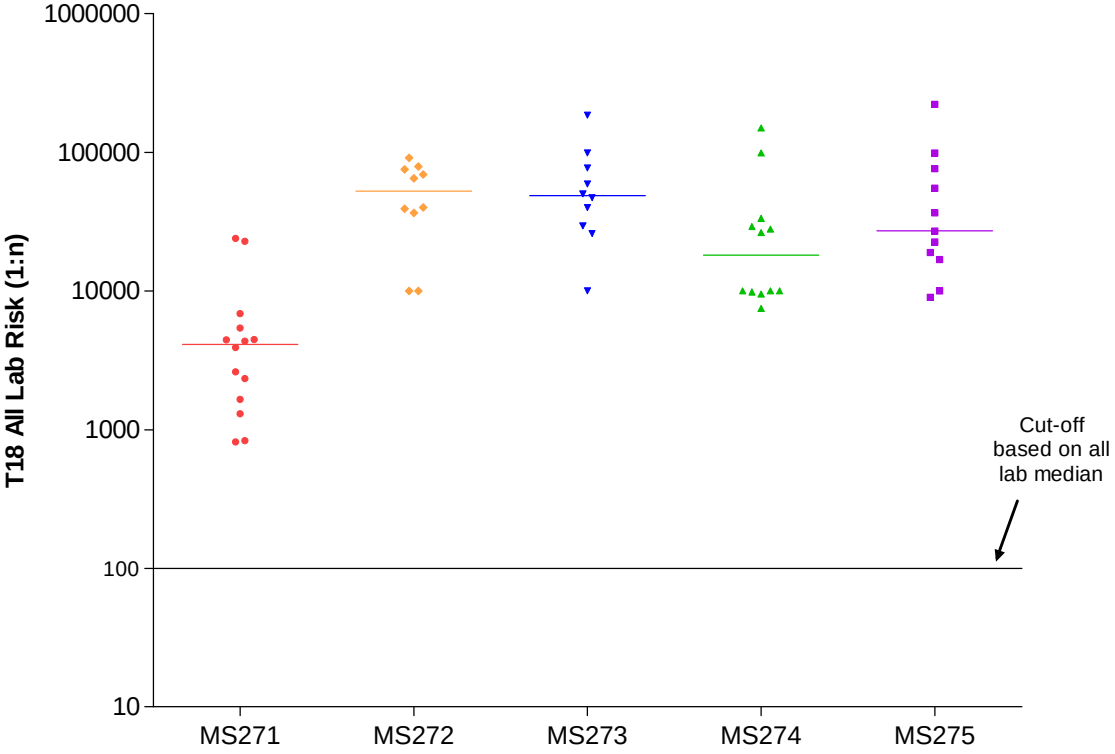


Figure 5

Graphic Distribution of Second Trimester Trisomy 21 Triple Risk Estimates

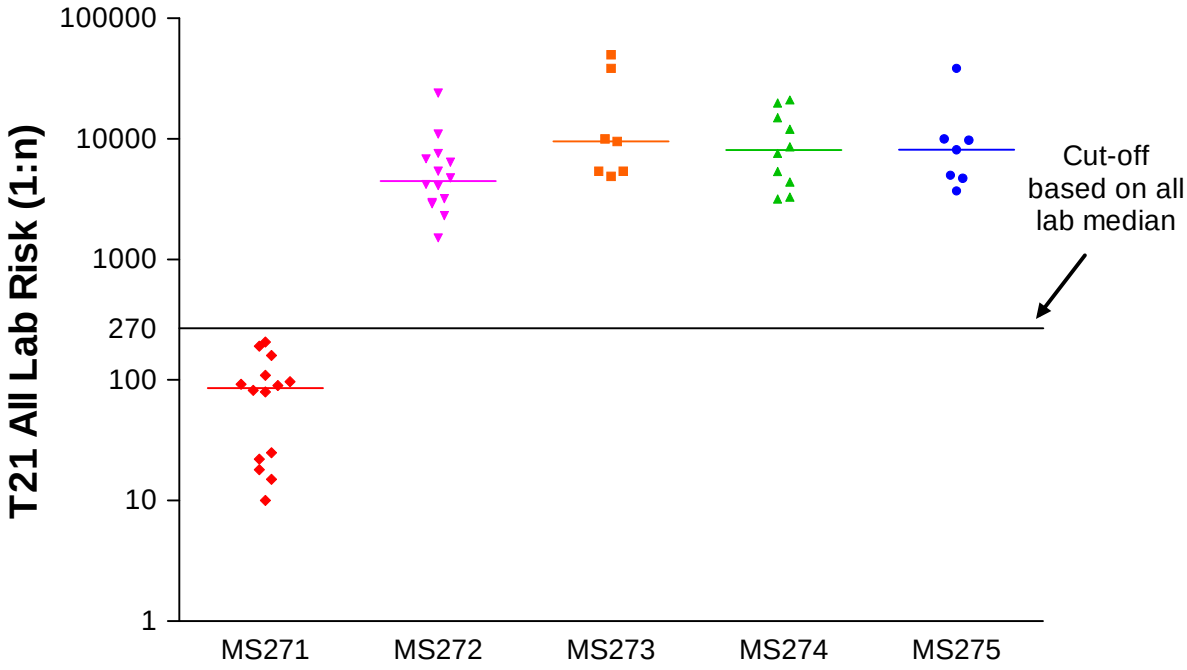


Figure 6

Graphic Distribution of Second Trimester Trisomy 21 Quadruple Risk Estimates

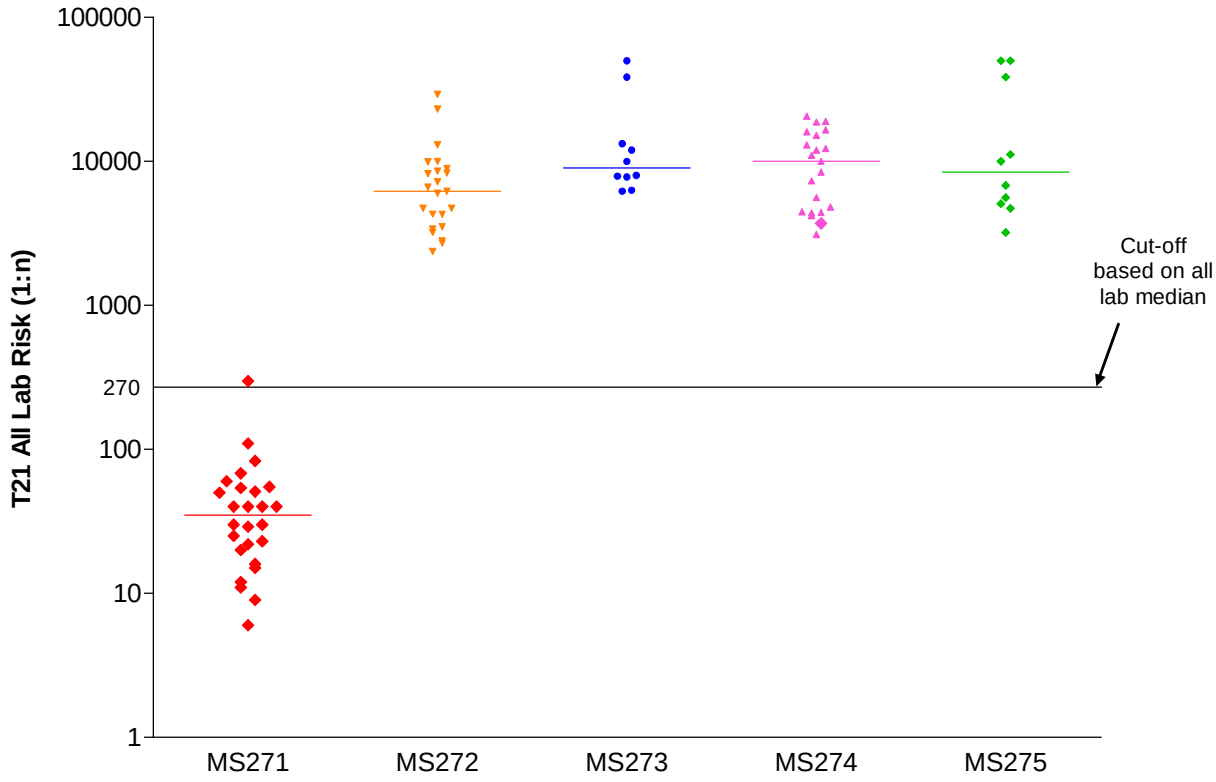


Figure 7A **MS AFP FEDM PT 9/11 Method Comparison**

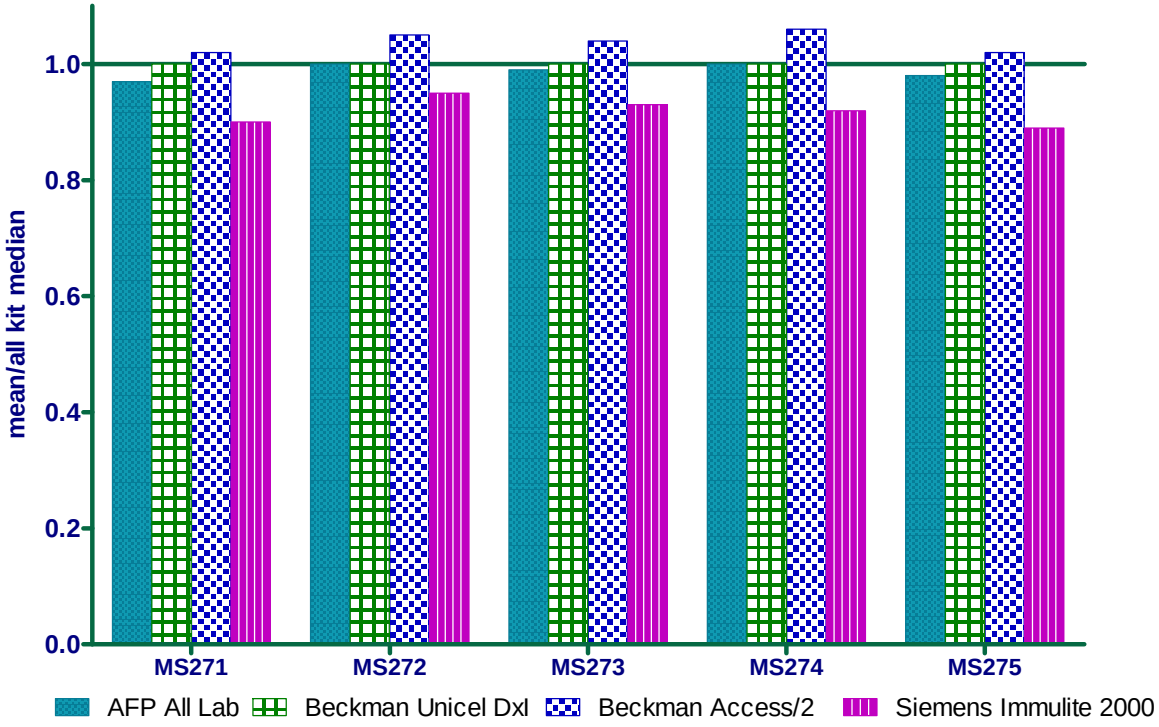


Figure 7B **AF AFP FEDM PT 9/11 Method Comparison**

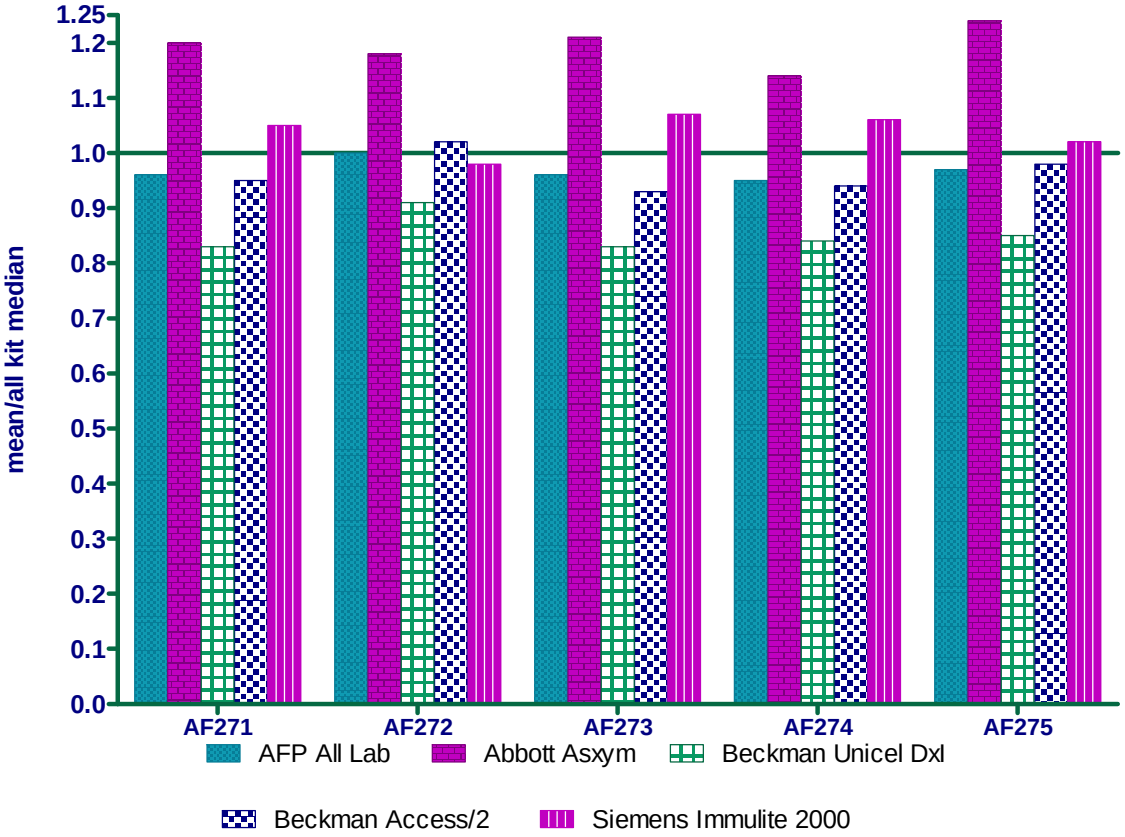


Figure 8A

MS uE3 FEDM PT 9/11 Method Comparison

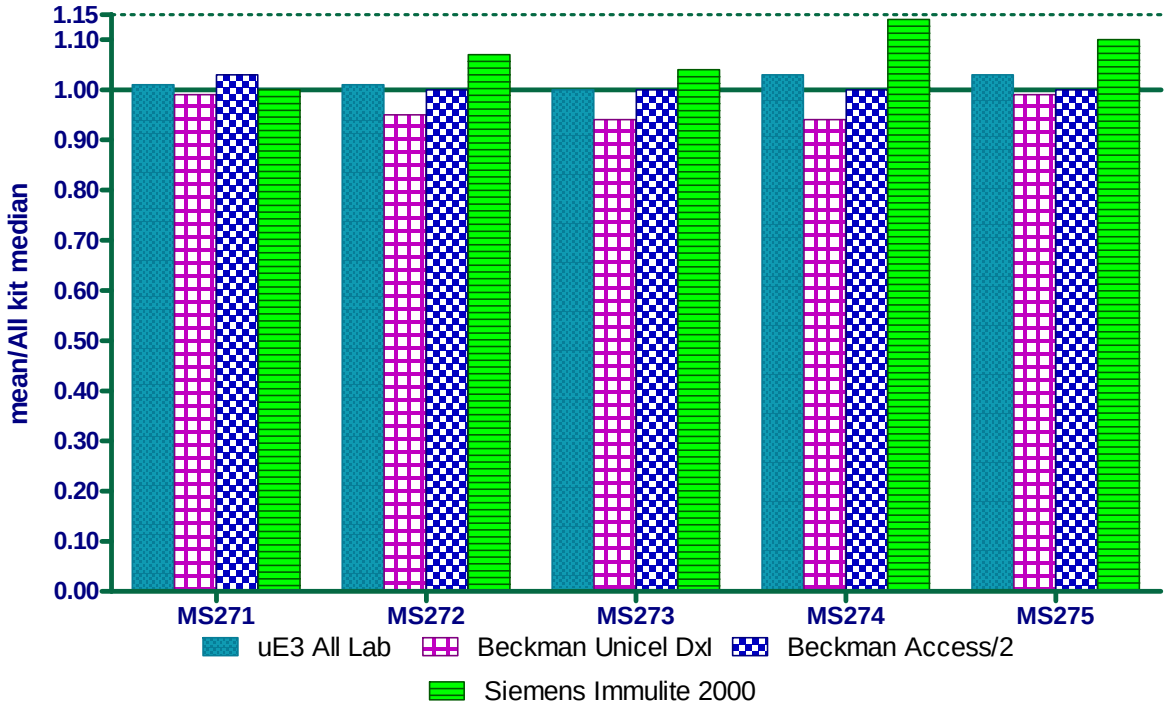


Figure 8 B

MS uE3 MOM FEDM PT 9/11 Method Comparison

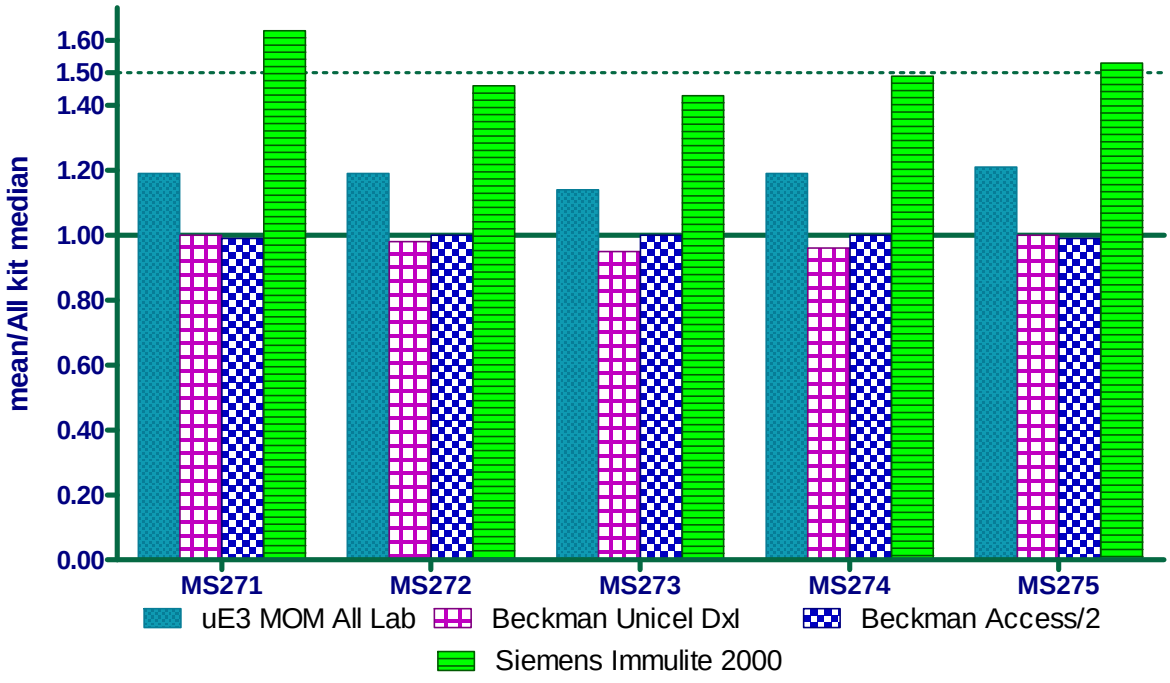


Figure 9

MS hCG FEDM PT 9/11 Method Comparison

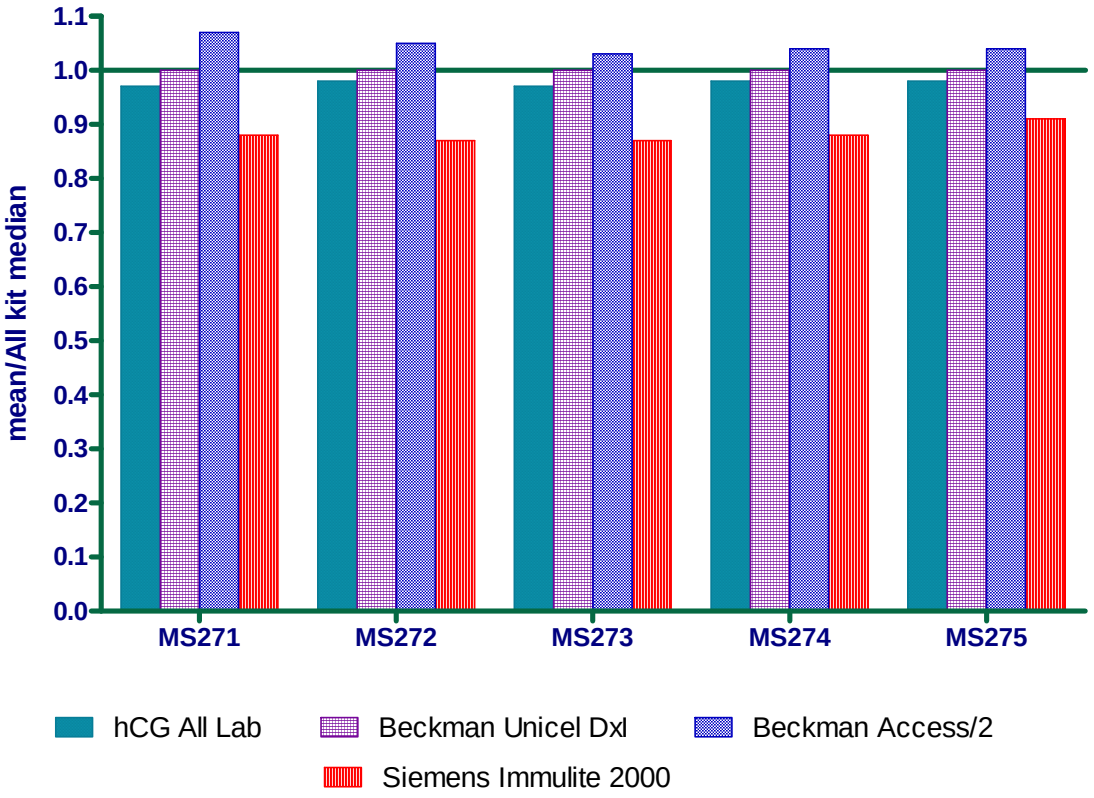


Figure 10

FT hCG FEDM PT 9/11 Method Comparison

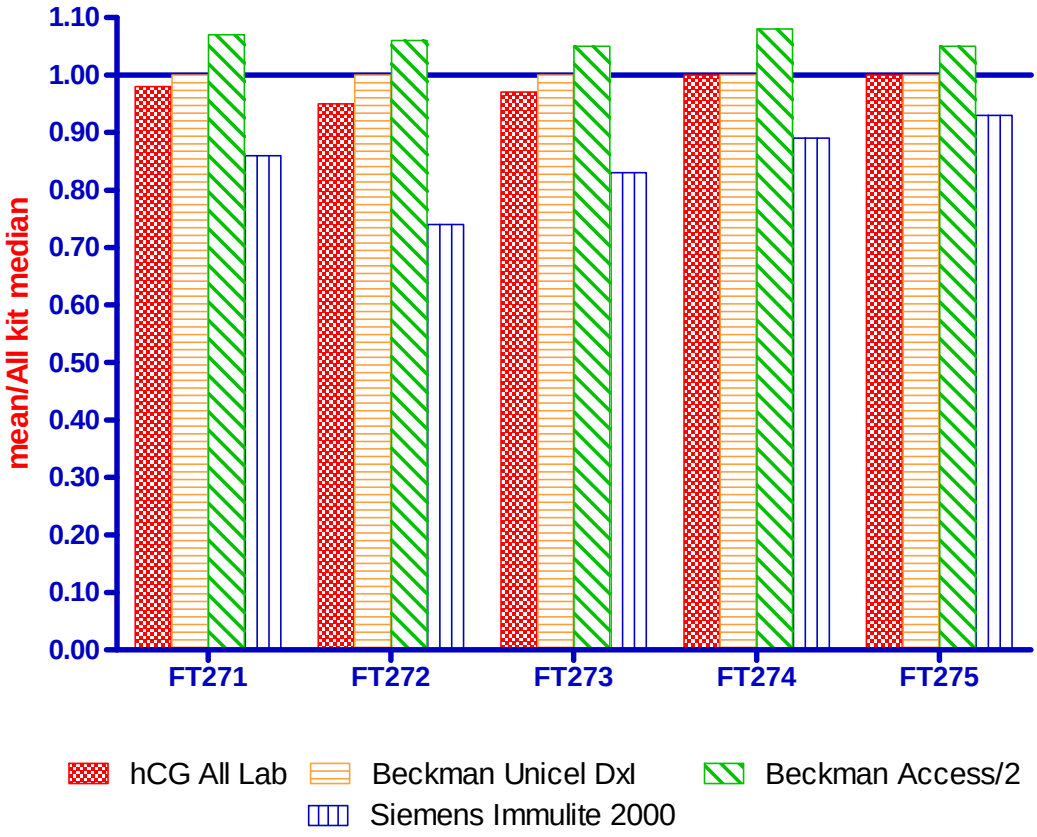


Figure 11A **MS Inhibin A FEDM PT 9/11 Method Comparison**

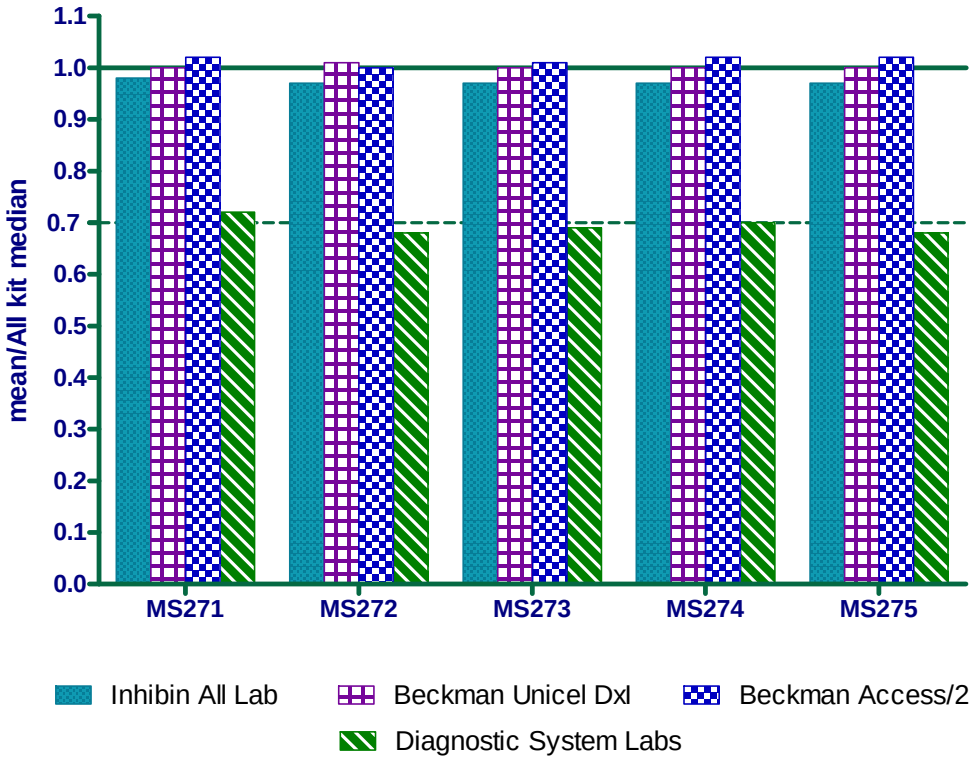


Figure 11B **MS Inhibin A MoM FEDM PT 9/11 Method Comparison**

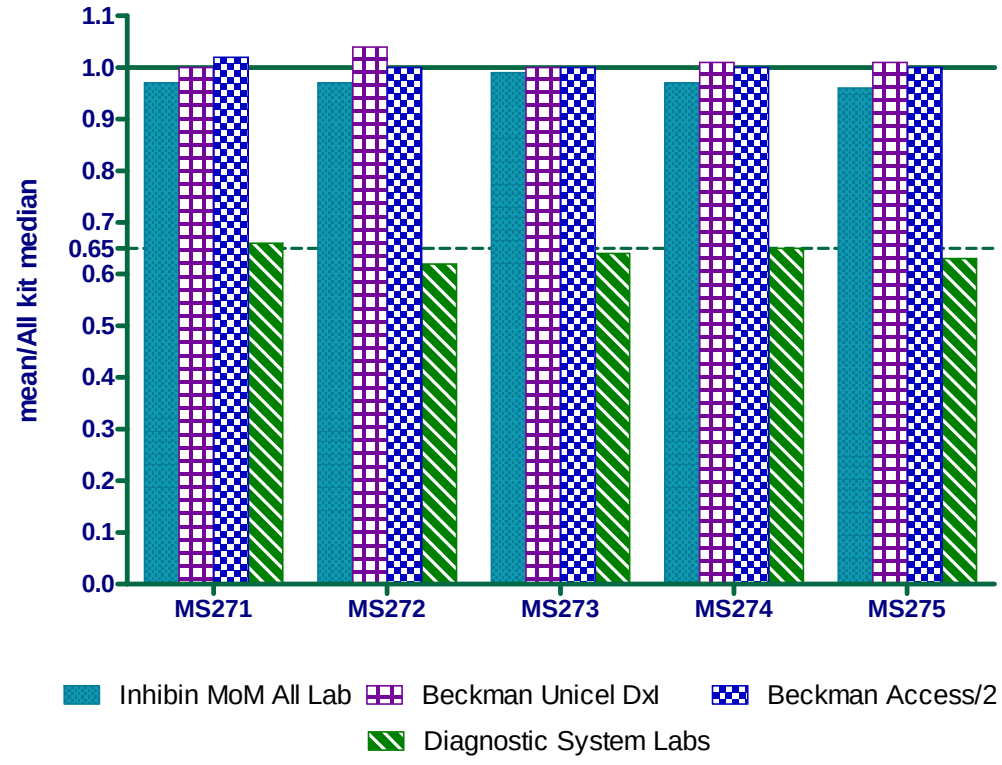
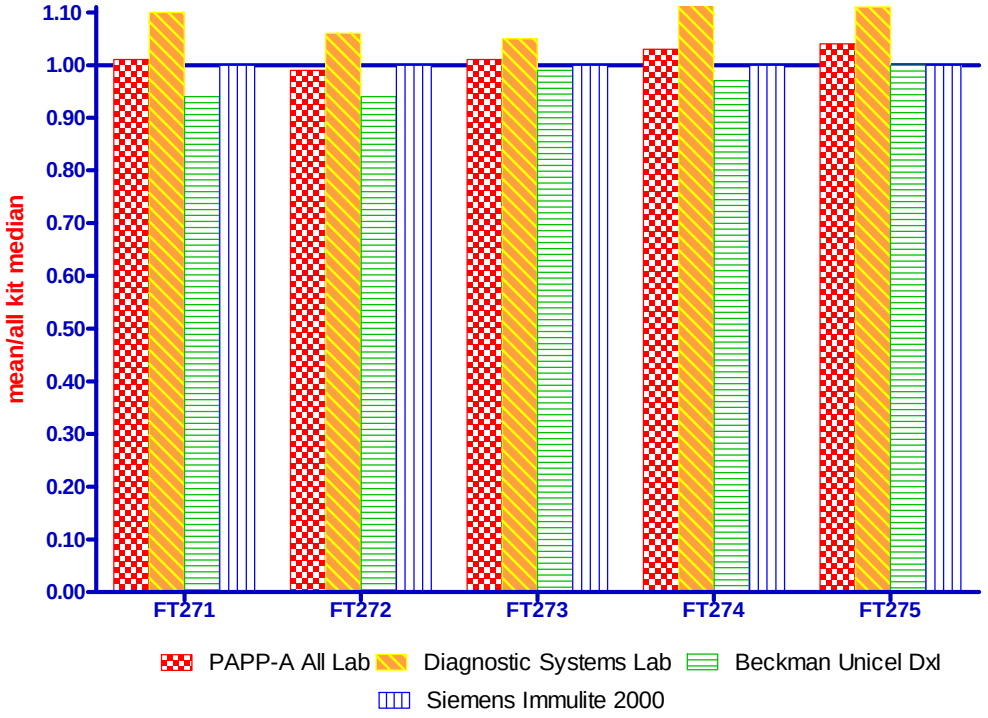


Figure 12A

FT PAPP- A FEDM PT 9/11 Method Comparison



*Please note: this graph is derived from converted values due to the difference in mass units used (mIU/ml -> ng/ml)

Figure 12B

FT PAPP- A MOM FEDM PT 9/11 Method Comparison

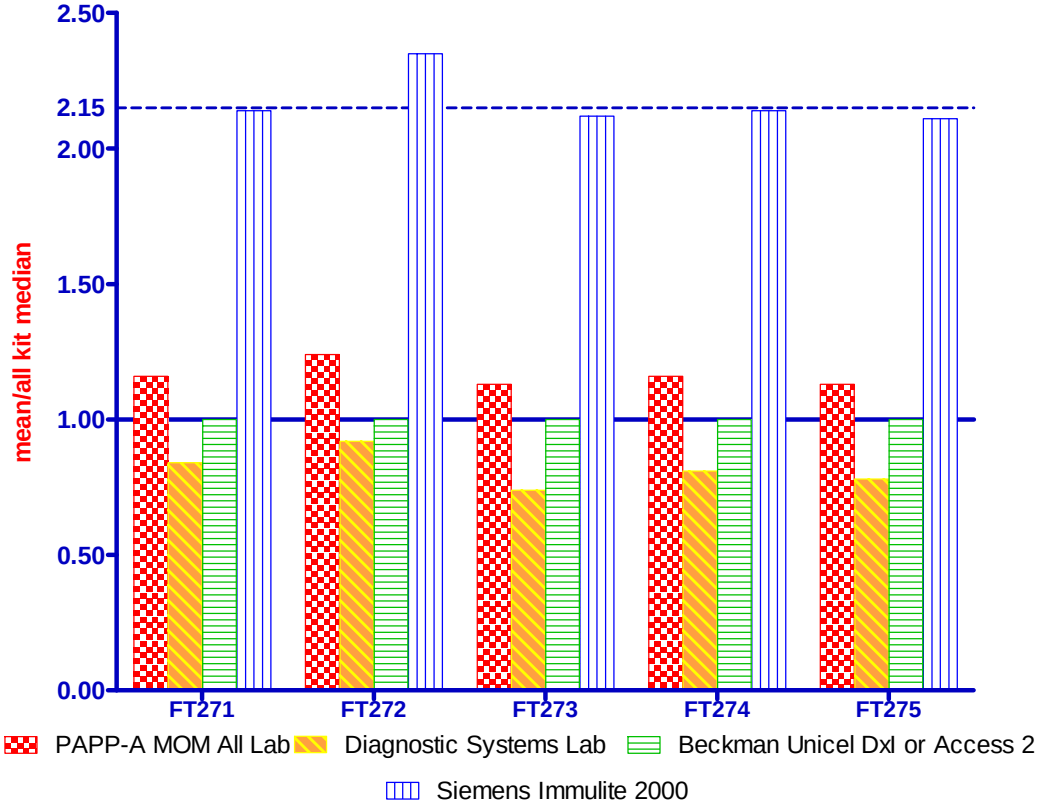


Figure 13

Graphic Distribution of First Trimester Trisomy 21 Risk Estimates

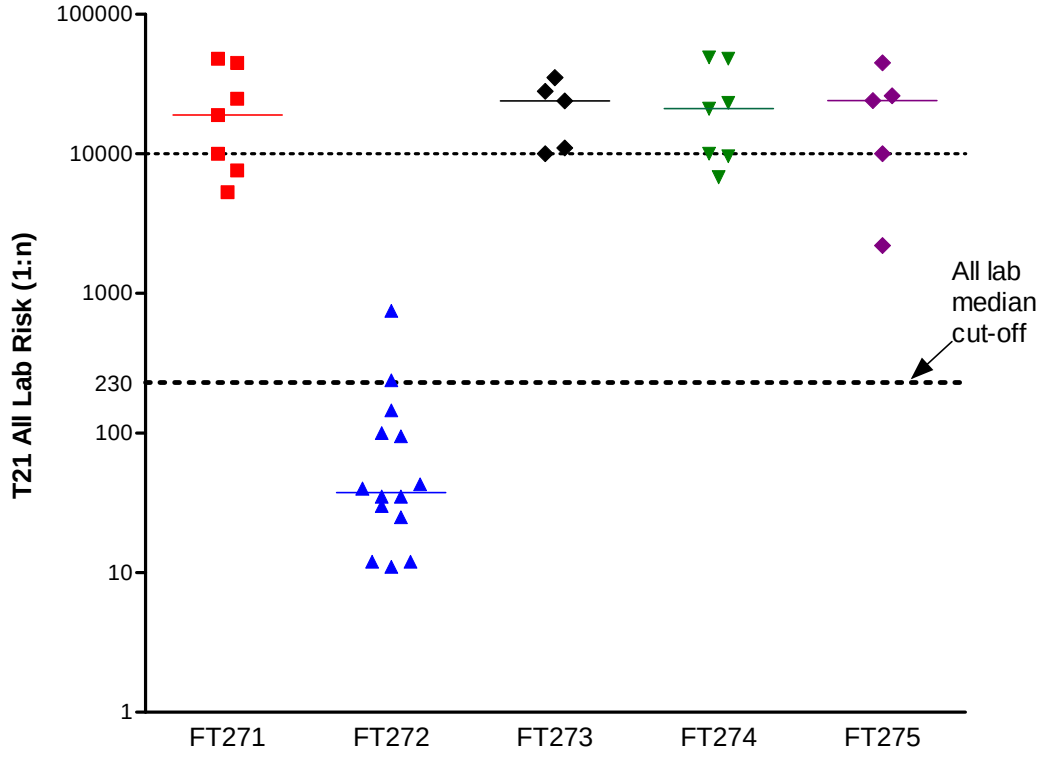
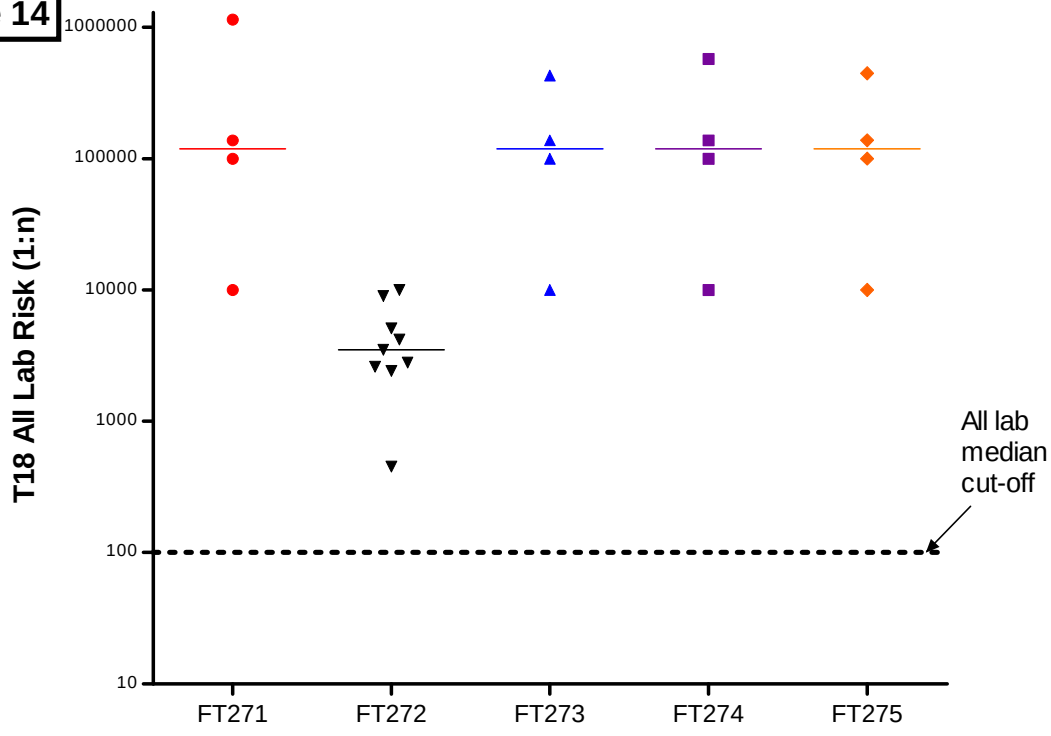


Figure 14

Graphic Distribution of First Trimester Trisomy 18 Risk Estimates



NEW YORK
state department of
HEALTH

Nirav R. Shah, M.D., M.P.H.
Commissioner

Sue Kelly
Executive Deputy Commissioner

Electronic Proficiency Test Reporting System Bulletin
September 2011

Laboratories participating in the September 2011 proficiency testing events in the categories listed below are required to submit results through the Electronic Proficiency Test Reporting System (EPTRS) system.

Bacteriology (Comprehensive, Gram Stains, Group A Streptococcus, Gonorrhea & Chlamydia, Throat Culture and Urine Culture)
Clinical Chemistry
Cytokines
Diagnostic Immunology
Endocrinology
Fetal Defect Markers
Human Immunodeficiency Virus
Mycology (C*ryptococcus neoformans* antigen, Identification - Molds, Identification -Yeasts and Susceptibility Testing)
Oncology Soluble Tumor Markers
Therapeutic Substance Monitoring/Quantitative Toxicology
Toxicology Blood Lead
Trace Elements (Serum, Urine and Whole Blood)
Virology (Comprehensive, HSV, Influenza, Rotavirus & RSV)

The Health Commerce System (HCS) Portal URL is https://commerce.health.state.ny.us
Please note: Version 3 of the Health Commerce System (HCS) was released July 27, 2011 and supports the use of Internet Explorer (IE) 7 and above. If you are using IE 6 to access the HCS, please insure that IE is upgraded on your computer.

After logging into the Portal, 'My Applications' is listed on the left side of the page. If you have access to EPTRS, the acronym 'EPTRS' will be listed under the heading 'My Applications'. Click on 'EPTRS' to access the homepage. If you do not see the acronym 'EPTRS', please send an email to clepeptrs@health.state.ny.us

Important Phone Numbers:

1. Technical Assistance with EPTRS - Monday through Friday between **8am and 4pm** by calling 518-486-5410.
2. Commerce Accounts Management Unit - for account information and passwords - Monday through Friday between 8am and 5 pm by calling 866-529-1890.

HCS Accounts – every user accessing EPTRS must have their own account for the Health Commerce System. It is a violation of the security and use agreement to share an account User ID and password with someone else. Sharing your account information with someone else will result in the suspension of your account.

EPTRS Webpages:

- Event Menu Page - Please review the laboratory's persistent data (instruments, reagents, methods, contact, email, etc). It is the responsibility of each laboratory to verify the data and make any required changes.
- Summary Page
    - Results submission - When you are ready to submit, navigate to the bottom of the Summary Page and click on the Submit/Attest button. Saving or validating without submitting results will result in a failure for non-participation. If you are unable to see the "Submit/Attest" button on the EPTRS Summary Page or if you have questions concerning result entry, please contact the Clinical Laboratory Evaluation Program at clepeptrs@health.state.ny.us.
    - Attestation statement - must be printed and signed by the laboratory director or responsible assistant director, the delegated submitter and the analyst prior to submission of the proficiency test results. The signed document must be kept on file in the laboratory for review by the laboratory surveyor during the next onsite survey.

If you experience any difficulty accessing EPTRS, please contact clepeptrs@health.state.ny.us

New York State Fetal Defect Markers Proficiency Test, FEDM PT, September 2011

PFI __ __ __ __ (1) **Lab Name and address** _____

Date samples obtained __ __ /__ __ /__ __ **Analyzed** __ __ / __ __ / __ __ (2) _____

Due Date: **September 28, 2011** _____

_____

Analyte Second Trimester Maternal Serum	**Analytical results**					Instrument code*	Reagent code*
	Vial **MS271**	Vial **MS272**	Vial **MS273**	Vial **MS274**	Vial **MS275**		
Gestational Age (weeks)	__ __.__ (3)	__ __.__ (4)	__ __.__ (5)	__ __.__ (6)	__ __.__ (7)		
MS AFP (ng/ml)	__ __ __.__ (8)	__ __ __.__ (9)	__ __ __.__ (10)	__ __ __.__ (11)	__ __ __.__ (12)	__ __ __ (13)	__ __ __ (14)
MS AFP MoM	__ __.__ __ (15)	__ __.__ __ (16)	__ __.__ __ (17)	__ __.__ __ (18)	__ __.__ __ (19)		
MS uE3 (ng/ml)	__ __.__ __ (20)	__ __.__ __ (21)	__ __.__ __ (22)	__ __.__ __ (23)	__ __.__ __ (24)	__ __ __ (25)	__ __ __ (26)
MS uE3 MoM	__ __.__ __ (27)	__ __.__ __ (28)	__ __.__ __ (29)	__ __.__ __ (30)	__ __.__ __ (31)		
MS hCG Please Check: _Total(IU/ml)/ _freeβ (mIU/ml)	__ __ __.__ (32)	__ __ __.__ (33)	__ __ __.__ (34)	__ __ __.__ (35)	__ __ __.__ (36)	__ __ __ (37)	__ __ __ (38)
MS hCG Total or Freeβ MoM	__ __.__ __ (39)	__ __.__ __ (40)	__ __.__ __ (41)	__ __.__ __ (42)	__ __.__ __ (43)		
MS Dimeric Inhibin A (pg/ml)	__ __ __.__ (44)	__ __ __.__ (45)	__ __ __.__ (46)	__ __ __.__ (47)	__ __ __.__ (48)	__ __ __ (49)	__ __ __ (50)
MS Dimeric Inhibin A MoM	__ __.__ __ (51)	__ __.__ __ (52)	__ __.__ __ (53)	__ __.__ __ (54)	__ __.__ __ (55)		
Neural Tube Screen 1 = positive, 0 = negative	__ (56)	__ (57)	__ (58)	__ (59)	__ (60)	NTD Based on: ❑ MoM cut-off ❑ Risk cut-off	←
Trisomy 21 Screen 1 = positive, 0 = negative	__ (61)	__ (62)	__ (63)	__ (64)	__ (65)	Based on: ❑ Quad ❑ Triple	←
Trisomy 18 Screen 1 = positive, 0 = negative	__ (66)	__ (67)	__ (68)	__ (69)	__ (70)		

New York State Fetal Defect Markers Proficiency Test, FEDM PT, September 2011

Amniotic Fluid	Vial **AF271**	Vial **AF272**	Vial **AF273**	Vial **AF274**	Vial **AF275**	Instrument code*	Reagent code*
AF AFP (μg/ml)	__ __ __.__ 71	__ __ __.__ 72	__ __ __.__ 73	__ __ __.__ 74	__ __ __.__ 75	___ ___ ___ 76	___ ___ ___ 77
AF AFP MoM	__ __.__ __ 78	__ __.__ __ 79	__ __.__ __ 80	__ __.__ __ 81	__ __.__ __ 82		
Interpretation **1** = **elevated w/ Ache indicated** **0 =Normal**	___ 83	___ 84	___ 85	___ 86	___ 87	Please indicate the Cut-off → MoM value used for interpretation	_____

***codes are on P. 4**

Risk Assessment Ratio (1:n) and Further Action	**MS271**	**MS272**	**MS273**	**MS274**	**MS275**	**Risk (MoM) Cut-off (white, Black, IDDM)**
NTD Risk (or MoM)						**White**_____ **Black**_____
R=Repeat, U=Ultrasound, A=Amnio NFA=NoFurtherAction, G=Genetic Counseling						**IDDM white**_____ **IDDM black**_____
Trisomy 21 Risk by Quad						**White**_____
R=Repeat, U=Ultrasound, A=Amnio NFA=NoFurtherAction, G=Genetic Counseling						**Black**_____ **IDDM**_____
Trisomy 21 Risk by Triple						**White**_____
R=Repeat, U=Ultrasound, A=Amnio NFA=NoFurtherAction, G=Genetic Counseling						**Black**_____ **IDDM**_____
Trisomy 18 Risk						**White**_____
R=Repeat, U=Ultrasound, A=Amnio NFA=NoFurtherAction, G=Genetic Counseling						**Black**_____ **IDDM**_____
Indicate software company used to calculate risk	_ αlpha	_ Benetech PRA		_ RMA	_other_____	

We, the undersigned, attest that the findings provided were produced in this laboratory from the analysis of proficiency test samples which were introduced into the routine workflow of the laboratory and analyzed using protocols and procedures which are (or which will be) routinely applied to **clinical** specimens. We further attest that the laboratory did not engage in any form of communication with individuals outside of our laboratory regarding the proficiency test and/or results obtained therefrom. **The laboratory director or the authorized assistant director who holds a CQ in Fetal Defect Markers must sign this form (stamps are not acceptable). If the director does not hold a CQ in this category, then the assistant director holding the appropriate CQ must sign. Do not forget to add your CQ codes; these are required for proper tracking of your results. Forms without all the required information will be returned. Failure to submit the required signatures will result in a score of zero.**

Analyst _____ **Laboratory director** _____ **CQ code**__ __ __ __ __ __

Analyst _____ **Assistant director** _____ **CQ code**__ __ __ __ __ __

(Please print and sign your names)

New York State Fetal Defect Markers Proficiency Test, FEDM PT, September 2011

Demographic Data:

Sample	Date of Birth	Race (B,W,H)	NT[1] (mm)	M. Wt (lbs)	LMP[3]	**CRL[4] (mm)**	US[2]/ Draw Date
FT 271	1/1/1982	H	1.08	160	6/20/2011	**48**	9/9/2011
FT 272	1/1/1986	W	2.90	150	6/17/2011	**53**	9/9/2011
FT 273	1/1/1990	A	1.10	100	6/24/2011	**45**	9/9/2011
FT 274	1/1/1985	H	1.40	140	6/13/2011	**59**	9/9/2011
FT 275	1/1/1992	W	1.55	130	6/10/2011	**69**	9/9/2011

[1]NT = Nuchal Translucency [2]US = Ultrasound [3]LMP = Last Menstrual Period [4]CRL = Crown Rump Length

First Trimester Maternal Serum	Vial **FT 271**	Vial **FT 272**	Vial **FT 273**	Vial **FT 274**	Vial **FT 275**	Instrument code*	Reagent code*
FT Gestational Age (weeks)	__ __.__ 88	__ __.__ 89	__ __.__ 90	__ __.__ 91	__ __.__ 92		
FT NT MoM	__.__ __ 93	__.__ __ 94	__.__ __ 95	__.__ __ 96	__.__ __ 97		
FT hCG Please Check: _Total(IU/ml)/ _freeβ (mIU/ml)	__ __ __.__ 98	__ __ __.__ 99	__ __ __.__ 100	__ __ __.__ 101	__ __ __.__ 102	__ __ __ 103	__ __ __ 104
FT hCG Total or Freeβ MoM	__ __.__ __ 105	__ __.__ __ 106	__ __.__ __ 107	__ __.__ __ 108	__ __.__ __ 109		
FT PAPP-A Please Check: _ mIU/ml _ng/ml	__ __.__ __ 110	__ __.__ __ 111	__ __.__ __ 112	__ __.__ __ 113	__ __.__ __ 114	__ __ __ 115	__ __ __ 116
FT PAPP-A MoM	__ __.__ __ 117	__ __.__ __ 118	__ __.__ __ 119	__ __.__ __ 120	__ __.__ __ 121		
FT Trisomy 21 Screen 1 = positive, 0 = negative	__ 122	__ 123	__ 124	__ 125	__ 126		
FT Trisomy 18 Screen 1 = positive, 0 = negative	__ 127	__ 128	__ 129	__ 130	__ 131		

Results will *not* be graded. Information will be used for future possible implementation.

Risk Assessment Ratio (1:n)and Further Action	**FT271**	**FT272**	**FT273**	**FT274**	**FT275**	**Risk Cut-off (white, Black, IDDM)**
Trisomy 21 Risk by First Trimester						**White**_____ **Black**_____ **IDDM**_____
R=Repeat, U=Ultrasound, A=Amnio, G=Genetic Counseling, C=CVS NFA=NoFurtherAction						
Trisomy 18 Risk by First Trimester						**White**_____ **Black**_____ **IDDM**_____
R=Repeat, U=Ultrasound, A=Amnio, G=Genetic Counseling NFA=NoFurtherAction						
Indicate software company used to calculate risk	_ αlpha	_ Benetech PRA		_ RMA	_other_____	

Instrument codes:

Abbott AxSym	ABB
Abbott Architect	ABH
Automatic (Robotic) Pipetting Station with or and Microplate Reader	APM
Bayer/Siemens Technicon Immuno-1	TNM
Siemens (Chiron) ACS-180	COS
Siemens ADVIA-Centaur	COB
Beckman Access/2	BCX
Beckman Unicel Dxl	BCU
Beckman Array	BCA
Siemens Diagnostic Dimension Rxl	DUD
Siemens Diagnostic MARK V with or and Microplate Reader	DPC
Qiagen Plato 3000 with or and Microplate Reader	QPM
Siemens Diagnostic Products Immulite	DPB
Siemens Diagnostic Products Immulite 2000	DPD
Siemens Diagnostic Products Immulite 2500	DPF
Trinity Biotech Nexgen	TBN
(DSL ELISA) with Microplate Reader	MPR
DSL Ario	DSA
DSL DSX with or and Microplate Reader	DSX
DSL Plato	DSP
UV/Vis Spectrophotometer	UVA
Gamma Counter	GAA
Rocket Immuno-Electrophoresis	RCE
P E Wallac Delfia	WAD
Analyzer/Instrument not shown, **specify on form**	ZZZ

Reagent/kit codes:

Abbott AFP Mono/Poly	AB1
Abbott AFP Mono/Mono	AB2
Abbott hCG	AB3
Abbott βhCG	AB4
Siemens (formerly Bayer)	BA1
Siemens (formerly Chiron)	CO1
Beckman Coulter	BC1
Siemens Diagnostic (Dade Behring)	DA1
Beckman Coulter, DSL ELISA (formerly Diagnostic Systems Lab EIA)	DS1
Diagnostic Systems Lab liquid RIA	DS2
Diagnostic Systems Lab solid RIA	DS3
DiaSorin-Clinical Assays	DC1
Siemens Diagnostic (DPC) Coat-A-Count	DP1
Siemens DPC Immulite, Immulite 2000 or Immulite 2500	DP5
New **Siemens DPC Immulite, Immulite 2000 or Immulite 2500 for uE3 only**	**DP6**
In-House	IH1
P E Wallac Delfia kit	PE1
Reagent/Kit not listed, **specify on form**	ZZZ

If an instrument and/or reagent you are using are not listed please provide us with the information, so that we can include it in the future. If you do not perform an assay leave the fields empty. No special codes are needed to indicate that an assay is not performed.

New York State Fetal Defect Markers Proficiency Test,
September 2011
Summary of Results

	MS 271	MS 272	MS 273	MS 274	MS 275
Gestational Age All Lab Mean:					
Mean	15.0	17.0	18.0	19.0	20.0
SD	0.00	0.00	0.00	0.00	0.00
%CV	0.0%	0.0%	0.0%	0.0%	0.0%
X+3*SD	15.0	17.0	18.0	19.0	20.0
X-3*SD	15.0	17.0	18.0	19.0	20.0
N	27	26	27	27	27

	MS 271	MS 272	MS 273	MS 274	MS 275
MS AFP All Lab Mean:					
mean	14.2	37.5	115.9	57.4	172.1
SD	1.2	3.0	8.9	5.4	17.1
%CV	8.1%	7.9%	7.7%	9.4%	9.9%
mean+3SD	17.6	46.5	142.6	73.6	223.3
mean-3SD	10.7	28.6	89.2	41.1	120.9
N	26	27	27	27	27
median	14	38.1	114.0	56.2	171.7
mean/all kit median	0.97	1.00	0.99	1.00	0.98

	MS 271	MS 272	MS 273	MS 274	MS 275
MS AFP DPC Immulite 2000 (DPD/DP5) mean:					
mean	13.0	35.6	108.5	53.0	156.6
SD	0.4	2.4	2.9	2.0	3.9
%CV	2.9%	6.7%	2.7%	3.8%	2.5%
mean+3SD	14.2	42.7	117.1	59.0	168.2
mean-3SD	11.9	28.5	99.9	47.0	145.1
N	8	8	8	8	8
median	13.2	34.9	108.0	52.7	156.5
mean/all kit median	0.90	0.95	0.93	0.92	0.89

	MS 271	MS 272	MS 273	MS 274	MS 275
MS AFP Beckman Unicel (BCU/BC1) mean:					
Mean	14.5	37.5	116.7	57.6	175.5
SD	0.7	3.0	11.7	5.4	17.9
%CV	5.1%	7.9%	10.0%	9.5%	10.2%
mean + 3SD	16.8	46.4	151.8	73.9	229.2
mean - 3SD	12.3	28.6	81.6	41.2	121.7
N	8	8	8	8	8
Median	14.1	38.4	119.9	58.3	177.4
mean/All kit median	1.00	1.00	1.00	1.00	1.00

	MS 271	MS 272	MS 273	MS 274	MS 275
MS AFP Beckman Access (BCX/BC1) mean:					
mean	14.9	39.3	121.7	60.8	179.5
SD	1.2	2.8	6.0	5.7	14.5
%CV	8.4%	7.2%	5.0%	9.4%	8.1%
mean+3SD	18.6	47.8	139.8	78.0	223.1
mean-3SD	11.1	30.8	103.5	43.7	135.9
N	9	9	9	9	9
median	15.1	40.0	123.5	61.7	178.0
mean/all kit median	1.02	1.05	1.04	1.06	1.02

	MS 271	MS 272	MS 273	MS 274	MS 275
MS AFP MoMs All Lab Mean:					
mean	0.49	1.18	2.55	0.97	2.97
SD	0.05	0.11	0.22	0.12	0.32
%CV	10.7%	9.2%	8.8%	11.9%	10.7%
mean+3SD	0.65	1.50	3.22	1.32	3.93
mean-3SD	0.33	0.85	1.87	0.63	2.02
N	27	27	27	27	27

	MS 271	MS 272	MS 273	MS 274	MS 275
MS AFP kit average:					
mean	14.1	37.5	115.6	57.1	170.5
SD	1.0	1.9	6.6	3.9	12.2
all kit median	14.5	37.5	116.7	57.6	175.5

New York State Fetal Defect Markers Proficiency Test,
September 2011
Summary of Results

	MS 271	MS 272	MS 273	MS 274	MS 275
MS uE3 All Lab Mean:					
mean	0.32	0.94	1.06	1.26	1.42
SD	0.04	0.08	0.07	0.12	0.15
%CV	10.9%	8.5%	7.0%	9.9%	10.2%
mean+3SD	0.43	1.17	1.29	1.64	1.86
mean-3SD	0.22	0.70	0.84	0.89	0.99
N	26	26	26	26	26
mean/all kit median	1.01	1.01	1.00	1.03	1.03

	MS 271	MS 272	MS 273	MS 274	MS 275
MS uE3 Beckman Unicel (BCU/BC1) mean:					
Mean	0.32	0.88	1.01	1.15	1.37
SD	0.04	0.09	0.06	0.06	0.09
%CV	12.1%	10.7%	5.6%	4.9%	6.8%
mean+3SD	0.43	1.17	1.18	1.32	1.65
mean-3SD	0.20	0.60	0.84	0.98	1.09
N	8	8	8	8	8
Median	0.32	0.92	1.00	1.15	1.41
mean/all kit median	0.99	0.95	0.94	0.94	0.99

	MS 271	MS 272	MS 273	MS 274	MS 275
MS uE3 BeckmanAccess (BCX/BC1) mean:					
mean	0.33	0.93	1.07	1.22	1.38
SD	0.02	0.05	0.06	0.06	0.08
%CV	5.0%	5.2%	5.2%	4.5%	5.6%
mean+3SD	0.38	1.07	1.23	1.39	1.61
mean-3SD	0.28	0.78	0.90	1.06	1.15
N	9	9	9	9	9
median	0.33	0.92	1.06	1.21	1.39
mean/all kit median	1.03	1.00	1.00	1.00	1.00

	MS 271	MS 272	MS 273	MS 274	MS 275
MS uE3 DPC Immulite 2000 (DPD/DP6) mean:					
Mean	0.32	0.99	1.11	1.40	1.52
SD	0.05	0.06	0.08	0.09	0.19
%CV	14.5%	6.2%	6.9%	6.6%	12.6%
mean+3SD	0.46	1.17	1.34	1.67	2.09
mean-3SD	0.18	0.80	0.88	1.12	0.94
N	9	9	9	9	9
Median	0.31	0.96	1.09	1.40	1.48
mean/All Kit Median	1.00	1.07	1.04	1.14	1.10

	MS 271	MS 272	MS 273	MS 274	MS 275
MS UE3 kit average:					
mean	0.32	0.93	1.06	1.26	1.42
SD	0.01	0.05	0.05	0.13	0.08
all kit median	0.32	0.93	1.07	1.22	1.38

New York State Fetal Defect Markers Proficiency Test,
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Summary of Results

	MS 271	MS 272	MS 273	MS 274	MS 275
MS uE3 MoMs All Lab Mean:					
Mean	0.58	1.14	0.93	0.86	0.86
SD	0.18	0.32	0.21	0.24	0.24
%CV	31.4%	28.1%	23.0%	27.6%	27.5%
X+3SD	1.12	2.10	1.57	1.58	1.57
X-3SD	0.03	0.18	0.29	0.15	0.15
N	26	27	26	27	27
mean/All Kit Median	1.19	1.19	1.14	1.19	1.21

	MS 271	MS 272	MS 273	MS 274	MS 275
MS uE3 MoMs (BCU/BC1) Mean:					
Mean	0.49	0.94	0.78	0.70	0.71
SD	0.08	0.11	0.06	0.07	0.03
%CV	16.0%	12.0%	7.4%	9.6%	4.6%
X+3SD	0.72	1.28	0.95	0.90	0.81
X-3SD	0.25	0.60	0.61	0.50	0.61
N	8	8	8	8	8
mean/All Kit Median	1.00	0.98	0.95	0.96	1.00

	MS 271	MS 272	MS 273	MS 274	MS 275
MS uE3 MoMs (BCX/BC1) Mean:					
Mean	0.48	0.96	0.82	0.72	0.70
SD	0.03	0.06	0.07	0.06	0.06
%CV	6.7%	5.9%	8.9%	8.9%	8.1%
X+3SD	0.58	1.13	1.03	0.91	0.88
X-3SD	0.39	0.79	0.60	0.53	0.53
N	9	9	9	9	9
mean/All Kit Median	0.99	1.00	1.00	1.00	0.99

	MS 271	MS 272	MS 273	MS 274	MS 275
MS uE3 MoM (DPD/DP6) Mean:					
Mean	0.79	1.39	1.17	1.08	1.09
SD	0.27	0.25	0.18	0.13	0.17
%CV	34.4%	17.6%	15.6%	12.3%	15.3%
X+3SD	1.61	2.13	1.71	1.47	1.59
X-3SD	-0.02	0.66	0.62	0.68	0.59
N	9	9	9	9	9
mean/All Kit Median	1.63	1.46	1.43	1.49	1.53

MS UE3 MoM kit average:					
mean	0.59	1.10	0.92	0.83	0.83
SD	0.18	0.26	0.21	0.21	0.22
all kit median	0.49	0.96	0.82	0.72	0.71

New York State Fetal Defect Markers Proficiency Test,
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Summary of Results

	MS 271	MS 272	MS 273	MS 274	MS 275
MS hCG All Lab Mean:					
mean	64.40	19.93	17.77	15.29	14.10
SD	8.35	1.91	1.77	1.45	1.25
%CV	13.0%	9.6%	10.0%	9.5%	8.9%
mean+3SD	89.5	25.7	23.1	19.6	17.9
mean-3SD	39.3	14.2	12.5	10.9	10.4
N	27	27	27	27	27
mean/all kit median	0.97	0.98	0.97	0.98	0.98

	MS 271	MS 272	MS 273	MS 274	MS 275
MS hCG DPC Immulite 2000 (DPD/DP5) mean:					
mean	55.9	17.7	15.9	13.8	13.0
SD	6.3	1.2	1.6	1.1	1.0
%CV	11.2%	6.5%	10.3%	8.1%	7.9%
mean+3SD	74.6	21.2	20.8	17.2	16.1
mean-3SD	37.1	14.3	11.0	10.5	9.9
N	8	8	8	8	8
median	54.7	17.9	15.5	13.6	12.8
mean/all kit median	0.85	0.87	0.87	0.88	0.91

	MS 271	MS 272	MS 273	MS 274	MS 275
MS hCG Beckman Unicel (BCU/BC1) mean:					
mean	66.10	20.43	18.36	15.64	14.33
SD	3.24	0.99	1.02	1.06	1.02
%CV	4.9%	4.8%	5.6%	6.8%	7.1%
mean+3SD	92.40	26.07	22.99	19.83	18.37
mean-3SD	49.36	16.62	14.68	12.68	11.34
N	8	8	8	8	8
median	66.45	20.35	18.25	15.70	14.15
mean/All kit median	1.00	1.00	1.00	1.00	1.00

	MS 271	MS 272	MS 273	MS 274	MS 275
MS hCG Beckman Access (BCX/BC1) mean:					
mean	70.9	21.3	18.8	16.3	14.9
SD	7.2	1.6	1.4	1.2	1.2
%CV	10.1%	7.4%	7.4%	7.3%	7.9%
mean+3SD	92.4	26.1	23.0	19.8	18.4
mean-3SD	49.4	16.6	14.7	12.7	11.3
N	9	9	9	9	9
median	71.3	21.3	18.7	15.8	14.7
mean/all kit median	1.07	1.05	1.03	1.04	1.04

	MS 271	MS 272	MS 273	MS 274	MS 275
MS hCG MoMs All Lab Mean:					
mean	1.71	1.00	0.86	0.74	0.85
SD	0.23	0.10	0.09	0.09	0.09
%CV	13.4%	9.9%	10.3%	12.3%	10.3%
mean+3SD	2.39	1.30	1.12	1.01	1.12
mean-3SD	1.02	0.71	0.59	0.46	0.59
N	26	26	26	26	26

	MS 271	MS 272	MS 273	MS 274	MS 275
MS hCG kit average:					
mean	64.3	19.8	17.7	15.2	14.1
SD	7.7	1.9	1.6	1.3	0.9
all kit median	66.1	20.4	18.4	15.6	14.3

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	MS 271	MS 272	MS 273	MS 274	MS 275
MS Inhibin A all lab mean:					
Mean	305.83	168.30	148.50	212.79	245.15
SD	36.65	20.24	17.82	25.61	31.16
%CV	12.0%	12.0%	12.0%	12.0%	12.7%
mean + 3SD	415.8	229.0	202.0	289.6	338.6
mean- 3SD	195.9	107.6	95.1	136.0	151.7
N	26	26	26	26	26
All Lab Median	317.7	173.9	153.4	219.7	253.4
mean/all kit median	0.98	0.97	0.97	0.97	0.97

	MS 271	MS 272	MS 273	MS 274	MS 275
MS Inhibin A Beckman Access (BCX/BC1) mean:					
Mean	318.8	174.3	155.1	222.7	257.3
SD	19.8	6.3	6.3	7.0	11.2
%CV	6.2%	3.6%	4.1%	3.1%	4.3%
mean + 3SD	378.1	193.0	174.1	243.5	290.8
mean- 3SD	259.4	155.5	136.1	201.8	223.7
N	13	13	13	13	13
median	329.5	173.7	155.1	223.8	255.0
mean/All kit median	1.02	1.00	1.01	1.02	1.02

	MS 271	MS 272	MS 273	MS 274	MS 275
MS Inhibin A Beckman Unicel (BCU/BC1) mean:					
Mean	312.8	175.4	153.0	218.3	251.7
SD	22.1	9.8	8.8	16.2	17.3
%CV	7.1%	5.6%	5.8%	7.4%	6.9%
mean + 3SD	379.0	204.7	179.5	267.0	303.6
mean- 3SD	246.6	146.1	126.5	169.5	199.7
N	10	10	10	10	10
median	320.7	179.2	149.7	216.7	253.0
mean/all kit median	1.00	1.01	1.00	1.00	1.00

	MS 271	MS 272	MS 273	MS 274	MS 275
MS Inhibin A Diagnostic System Labs (DS1) Mean:					
Mean	226.4	118.7	104.9	151.8	170.8
SD	38.1	16.9	12.1	18.7	24.2
%CV	16.8%	14.3%	11.5%	12.3%	14.1%
mean + 3SD	340.7	169.6	141.1	207.8	243.3
mean- 3SD	112.1	67.9	68.8	95.7	98.4
N	3	3	3	3	3
median	211.5	109.0	104.7	146.9	158.0
mean/all kit median	0.72	0.68	0.69	0.70	0.68

	MS 271	MS 272	MS 273	MS 274	MS 275
MS Inhibin A kit average:					
mean	286.0	156.1	137.7	197.6	226.6
SD	51.7	32.4	28.4	39.7	48.4
all kit median	312.8	174.3	153.0	218.3	251.7

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Summary of Results

	MS 271	MS 272	MS 273	MS 274	MS 275
MS Inhibin A MoM All Lab Mean:					
mean	1.63	1.13	0.89	1.11	1.29
SD	0.24	0.17	0.08	0.18	0.19
%CV	14.9%	15.0%	8.7%	16.5%	14.9%
mean+3SD	2.35	1.63	1.12	1.66	1.87
mean-3SD	0.90	0.62	0.66	0.56	0.71
N	26	26	24	26	26
mean/All kit median	0.97	0.97	0.99	0.97	0.96

	MS 271	MS 272	MS 273	MS 274	MS 275
MS Inhibin A MoM Beckman Access (BCX/BC1) mean:					
Mean	1.71	1.16	0.90	1.15	1.35
SD	0.17	0.06	0.07	0.10	0.11
%CV	9.8%	5.4%	7.3%	8.8%	7.8%
X + 3SD	2.21	1.35	1.09	1.46	1.66
X - 3SD	1.21	0.97	0.70	0.85	1.03
N	13	13	13	13	13
Kit Median	1.71	1.17	0.89	1.13	1.35
mean/All kit median	1.02	1.00	1.00	1.00	1.00

	MS 271	MS 272	MS 273	MS 274	MS 275
MS Inhibin A MoM Beckman Unicel (BCU/BC1) mean:					
Mean	1.67	1.21	0.90	1.17	1.36
SD	0.15	0.09	0.07	0.16	0.10
%CV	8.7%	7.8%	7.5%	13.5%	7.2%
X + 3SD	2.11	1.49	1.10	1.64	1.65
X - 3SD	1.24	0.92	0.70	0.69	1.07
N	10	10	10	10	10
Kit Median	1.66	1.16	0.89	1.09	1.30
mean/All kit median	1.00	1.04	1.00	1.01	1.01

	MS 271	MS 272	MS 273	MS 274	MS 275
MS Inhibin A MoM Diagnostic System Labs (DS1) Mean:					
Mean	1.11	0.72	0.58	0.75	0.84
SD	0.07	0.06	0.11	0.17	0.11
%CV	6.0%	7.7%	18.2%	22.1%	12.6%
X + 3SD	1.31	0.88	0.89	1.25	1.16
X - 3SD	0.91	0.55	0.26	0.25	0.53
N	3	3	3	3	3
Kit Median	1.14	0.69	0.58	0.73	0.86
mean/All kit median	0.66	0.62	0.64	0.65	0.63

	MS 271	MS 272	MS 273	MS 274	MS 275
MS Inhibin A MoM kit average:					
mean	1.5	1.0	0.8	1.0	1.2
SD	0.3	0.3	0.2	0.2	0.3
all kit median	1.7	1.2	0.9	1.2	1.3

New York State Fetal Defect Markers Proficiency Test,
September 2011
Summary of Results

	AF 271	AF 272	AF 273	AF 274	AF 275
AF AFP All Lab Mean :					
mean	9.29	4.56	7.14	6.00	18.71
SD	1.50	0.57	1.29	0.88	2.88
%CV	16.2%	12.5%	18.1%	14.7%	15.4%
mean+3SD	13.8	6.3	11.0	8.6	27.4
mean-3SD	4.8	2.9	3.3	3.4	10.1
N	22	22	22	22	22
All kit median	9.7	4.6	7.5	6.3	19.2
mean/All kit mean	0.96	1.00	0.96	0.95	0.97

	AF 271	AF 272	AF 273	AF 274	AF 275
AF AFP DPC Immulite 2000 (DPD/DP5) mean:					
mean	10.1	4.5	8.0	6.7	19.6
SD	1.1	0.4	0.9	0.8	1.1
%CV	10.8%	9.7%	11.5%	11.4%	5.7%
mean+3SD	13.4	5.8	10.7	9.0	22.9
mean-3SD	6.8	3.2	5.2	4.4	16.3
N	5	5	5	5	5
median	10.6	4.4	7.7	6.8	19.2
mean/all kit median	1.05	0.98	1.07	1.06	1.02

	AF 271	AF 272	AF 273	AF 274	AF 275
AF AFP MoMs All Lab Mean:					
mean	0.53	0.57	1.09	1.04	2.87
SD	0.06	0.05	0.15	0.14	0.32
%CV	10.4%	9.1%	13.8%	13.6%	11.1%
mean+3SD	0.70	0.73	1.54	1.47	3.82
mean-3SD	0.37	0.41	0.64	0.62	1.91
N	21	21	21	21	21

	AF 271	AF 272	AF 273	AF 274	AF 275
AF AFP Beckman Unicel (BCU/BC1) mean:					
Mean	8.0	4.2	6.2	5.3	16.4
SD	1.1	0.4	0.5	0.6	2.2
%CV	13.1%	8.4%	8.2%	11.2%	13.6%
X+3SD	12.7	6.4	10.9	8.1	25.9
X-3SD	5.7	2.9	3.0	3.8	11.8
N	7	7	7	7	7
median	8.1	4.1	6.2	5.0	15.5
mean/All kit median	0.83	0.91	0.83	0.84	0.85

	AF 271	AF 272	AF 273	AF 274	AF 275
AF AFP Beckman Access (BCX/BC1) mean:					
mean	9.2	4.7	6.9	5.9	18.9
SD	1.2	0.6	1.3	0.7	2.3
%CV	12.7%	12.3%	18.9%	12.1%	12.4%
mean+3SD	12.7	6.4	10.9	8.1	25.9
mean-3SD	5.7	2.9	3.0	3.8	11.8
N	7	7	7	7	7
median	8.7	4.5	7.1	5.9	18.6
mean/all kit median	0.95	1.02	0.93	0.94	0.98

	AF 271	AF 272	AF 273	AF 274	AF 275
AF AFP Abbott Axsym (ABB/AB2) mear:					
mean	11.6	5.4	9.1	7.2	23.9
N	2	2	2	2	2
mean/all kit median	1.20	1.18	1.21	1.14	1.24

	AF 271	AF 272	AF 273	AF 274	AF 275
AF AFP kit average:					
mean	9.7	4.7	7.5	6.3	19.7
SD	1.5	0.5	1.2	0.8	3.1
all kit median	9.7	4.6	7.5	6.3	19.2

New York State Fetal Defect Markers Proficiency Test,
September 2011
Summary of First Trimester Results

	FT271	FT272	FT273	FT274	FT275
FT Gestational Age All Lab Mean:					
Mean	11.5	11.9	11.2	12.4	13.1
SD	0.13	0.11	0.13	0.09	0.06
%CV	1.1%	0.9%	1.2%	0.7%	0.5%
X+3*SD	11.9	12.2	11.6	12.6	13.2
X-3*SD	11.1	11.6	10.8	12.1	12.9
N	17	17	17	17	17

	FT271	FT272	FT273	FT274	FT275
FT NT MoMs All Lab Mean:					
Mean	0.89	2.17	0.95	0.95	0.93
SD	0.05	0.16	0.06	0.06	0.06
%CV	6.1%	7.3%	6.7%	6.4%	6.5%
X+3SD	1.05	2.65	1.14	1.13	1.11
X- 3SD	0.72	1.70	0.76	0.77	0.75
N	16	16	16	16	16
All Median	0.89	2.13	0.95	0.94	0.93

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	FT271	FT272	FT273	FT274	FT275
FT hCG All Lab Mean:					
mean	70.09	148.04	69.56	59.49	56.62
SD	9.08	27.36	9.59	6.88	5.63
%CV	13.0%	18.5%	13.8%	11.6%	9.9%
X+3SD	97.3	230.1	98.3	80.1	73.5
X-3SD	42.9	66.0	40.8	38.8	39.7
N	16	16	16	16	16
mean/All kit median	0.98	0.95	0.97	1.00	1.00

	FT271	FT272	FT273	FT274	FT275
FT hCG Beckman Unicel (BCU/BC1) mean:					
mean	71.18	156.55	71.70	59.55	56.60
SD	2.70	12.83	9.08	3.58	2.58
%CV	3.8%	8.2%	12.7%	6.0%	4.6%
X+3SD	99.85	219.73	94.78	80.98	76.26
X-3SD	51.83	113.67	56.36	47.51	42.80
N	4	4	4	4	4
median	71.45	159.10	72.80	59.20	57.30
mean/All kit median	1.00	1.00	1.00	1.00	1.00

	FT271	FT272	FT273	FT274	FT275
FT hCG DPC Immulite 2000(DPD/DP5) mean:					
mean	61.2	115.1	59.4	52.8	52.6
SD	7.0	13.3	5.1	5.1	5.6
%CV	11.5%	11.6%	8.5%	9.7%	10.7%
X+3SD	82.2	155.1	74.6	68.1	69.5
X-3SD	40.1	75.1	44.3	37.5	35.6
N	5	5	5	5	5
median	62.7	120.3	60.7	55.8	50.7
mean/All kit median	0.86	0.74	0.83	0.89	0.93

	FT271	FT272	FT273	FT274	FT275
FT hCG Beckman Access (BCX/BC1) mean:					
mean	75.8	166.7	75.6	64.2	59.5
SD	8.0	17.7	6.4	5.6	5.6
%CV	10.6%	10.6%	8.5%	8.7%	9.4%
X+3SD	99.9	219.7	94.8	81.0	76.3
X-3SD	51.8	113.7	56.4	47.5	42.8
N	7	7	7	7	7
median	76.3	169.5	74.7	63.0	59.1
mean/All kit median	1.07	1.06	1.05	1.08	1.05

	FT271	FT272	FT273	FT274	FT275
FT hCG MoMs All Lab Mean:					
Mean	0.95	1.99	0.72	0.83	0.82
SD	0.11	0.23	0.12	0.08	0.10
%CV	11.4%	11.8%	17.3%	9.4%	12.7%
mean+3*SD	1.27	2.70	1.09	1.06	1.13
mean- 3*SD	0.63	1.29	0.35	0.59	0.51
N	15	15	15	15	15
All Median	0.94	2.01	0.71	0.83	0.81

	FT271	FT272	FT273	FT274	FT275
FT hCG kit average:					
mean	69.4	146.1	68.9	58.9	56.2
SD	7.5	27.3	8.4	5.8	3.5
all kit median	71.2	156.6	71.7	59.6	56.6

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	FT271	FT272	FT273	FT274	FT275
FT PAPP-A All Lab Mean:					
Mean	1629.23	798.22	1407.35	2055.52	2307.52
SD	312.86	103.63	171.21	412.36	346.91
%CV	19.2%	13.0%	12.2%	20.1%	15.0%
mean + 3SD	2567.81	1109.12	1920.99	3292.60	3348.24
mean- 3SD	690.65	487.32	893.70	818.44	1266.80
N	14	14	14	14	14
All Lab Median	1583.02	785.54	1382.82	1990.99	2245.38
mean/All kit median	1.01	0.99	1.01	1.03	1.04

	FT271	FT272	FT273	FT274	FT275
FT PAPP-A Beckman Unicel(BCU/BC1) Mean:					
Mean	1518.40	752.82	1370.42	1935.87	2218.93
SD	124.94	48.47	130.02	176.10	219.31
%CV	8.2%	6.4%	9.5%	9.1%	9.9%
X + 3SD	1893.22	898.24	1760.48	2464.16	2876.86
X - 3SD	1143.58	607.40	980.35	1407.58	1561.01
N	6	6	6	6	6
Kit Median	1521.0	770.5	1339.5	1977.5	2218.2
mean/All kit median	0.94	0.94	0.99	0.97	1.00

	FT271	FT272	FT273	FT274	FT275
FT PAPP-A kit average:					
mean	1634.22	802.00	1407.30	2055.52	2300.15
SD	128.84	48.28	49.02	158.25	150.27
all kit median	1611.26	803.84	1388.56	1995.75	2218.93

	FT271	FT272	FT273	FT274	FT275
FT PAPP-A DPC Immullite 2000 (DPD/DP5) Mean:					
Mean	1611.26	803.84	1388.56	1995.75	2207.96
SD	104.86	82.38	12.02	141.59	207.88
%CV	6.5%	10.2%	0.9%	7.1%	9.4%
X + 3SD	1925.84	1050.97	1424.60	2420.50	2831.62
X - 3SD	1296.68	556.71	1352.51	1570.99	1584.31
N	3	3	3	3	3
Kit Median	1611.26	821.81	1389.56	2045.68	2240.42
mean/All kit median	1.00	1.00	1.00	1.00	1.00

	FT271	FT272	FT273	FT274	FT275
***FT PAPP-A Diagnostic Systems Lab (DS1) Mean:**					
Mean	1773.00	849.33	1462.93	2234.96	2473.55
SD	498.60	148.97	260.56	662.91	505.70
%CV	28.1%	17.5%	17.8%	29.7%	20.4%
X + 3SD	4.71	2.31	3.57	5.62	5.35
X - 3SD	1.09	0.75	1.43	1.29	2.13
N	5	5	5	5	5
Kit Median	1586.04	817.55	1421.17	1956.98	2250.35
mean/All kit median	1.10	1.06	1.05	1.12	1.11

***Note: The above 2 tables contain converted values (mIU/ml->ng/ml) from equations obtained based on in house correlation data.**
(see critique)

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	FT271	FT272	FT273	FT274	FT275
FT PAPP-A MoM All Lab Mean:					
Mean	3.24	1.36	2.05	2.51	2.01
SD	1.60	0.68	1.04	1.22	1.03
%CV	49.3%	50.1%	51.0%	48.5%	51.3%
mean + 3SD	8.03	3.41	5.18	6.15	5.11
mean- 3SD	-1.55	-0.68	-1.08	-1.14	-1.08
N	14	14	14	14	14
All Lab Median	2.71	1.12	1.66	2.04	1.69
mean/ All kit median	1.16	1.24	1.13	1.16	1.13

	FT271	FT272	FT273	FT274	FT275
FT PAPP-A MoM DPC Immulite 2000 (DPD/DP5) Mean:					
Mean	5.97	2.59	3.84	4.65	3.74
SD	0.97	0.14	0.34	0.19	0.62
%CV	16.3%	5.3%	8.9%	4.0%	16.5%
X + 3SD	8.88	3.00	4.86	5.20	5.59
X - 3SD	3.06	2.18	2.82	4.09	1.89
N	3	3	3	3	3
mean/All kit median	2.14	2.35	2.12	2.14	2.11

	FT271	FT272	FT273	FT274	FT275
FT PAPP-A MoM Beckman Unicel(BCU/BC1) Mean:					
Mean	2.80	1.10	1.81	2.17	1.78
SD	0.28	0.09	0.37	0.28	0.37
%CV	10.0%	8.6%	20.6%	12.8%	20.7%
X + 3SD	3.63	1.39	2.93	3.00	2.88
X - 3SD	1.96	0.82	0.69	1.34	0.67
N	6	6	6	6	6
Kit Median	2.83	1.14	1.69	2.18	1.70
mean/All kit median	1.00	1.00	1.00	1.00	1.00

	FT271	FT272	FT273	FT274	FT275
FT PAPP-A MoM Diagnostic System Labs (DS1) Mean:					
Mean	2.36	1.01	1.34	1.76	1.39
SD	0.21	0.07	0.16	0.17	0.10
%CV	8.9%	6.7%	12.2%	9.6%	7.1%
X + 3SD	2.98	1.21	1.83	2.26	1.68
X - 3SD	1.73	0.81	0.85	1.25	1.09
N	4	4	4	4	4
Kit Median	2.31	0.99	1.31	1.75	1.41
mean/ All kit median	0.84	0.92	0.74	0.81	0.78

	FT271	FT272	FT273	FT274	FT275
FT PAPP-A MoM kit average:					
mean	3.71	1.57	2.33	2.86	2.30
SD	1.97	0.89	1.33	1.56	1.26
all kit median	2.80	1.10	1.81	2.17	1.78