NEW YORK
state department of
HEALTH

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Acting Commissioner of Health

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Executive Deputy Commissioner

Molecular and Cellular Tumor Marker Proficiency Test Event
MCTM 3-2014
Summary of results[1]

May 20, 2014

Dear Laboratory Director,

Below is a summary and discussion of the New York State Molecular and Cellular Tumor Markers proficiency test event MCTM 3-2014 from March 18, 2014, due date April 16, 2014.

Samples: All laboratories received three (3) different specimens prepared by Wadsworth Center personnel.

Evaluation: Laboratories were asked to perform those molecular assays for which they hold or have applied for a NYS permit. A total of 38 laboratories participated, performing various numbers and combinations of tests. The attached tables summarize the results and methods that were used by participating laboratories. In Table 1, a consensus interpretation is shown of **R**: rearranged/clonal band detected; **G**: germline/no clonal band detected; **WT**: wild-type; **MUT**: mutated; **NEG**: negative or not detected; **POS**: positive or detected; **O**: oligoclonal; **N**: no clonal band or fusion product detected. For IGHV only: **H**: clonal band detected and hypermutated; **U**: clonal band detected, but not hypermutated; **I** (Indeterminate) is shown if no consensus was reached because less than three labs performed a test, or if the concordance between labs was less than 80%. Please note that only the all method consensus is shown. If there were distinct method specific discrepancies these are discussed in the relevant section below.

Each lab will receive a personalized result sheet by regular mail that shows your lab's results in comparison to the all lab consensus (if any) derived from all methods combined. Two scores were calculated, one for each genotypic marker (**assay score**) across all three samples, and one for each sample (**sample score**) across all assays performed by your lab for each sample. From the latter we also calculated an overall score. Your **assay** score is expressed as a fraction, whereby the denominator is the number of samples you analyzed with a given assay and that were evaluable, i.e. produced a consensus, and the numerator is the number of samples for which you agreed with the consensus. For example, 3/3 means you analyzed all 3 samples and agreed with the consensus for all 3 of them. 1/2 would mean you analyzed only two samples or only 2 samples produced a consensus, but agreed with the consensus for only one of them. The assay score is indicated in the 'score' column to the right of each assay you performed. The **sample** score was calculated as the percentage of 'correct' answers per sample (i.e. that agree with the

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

consensus), based on the number of assays performed per sample by your lab that were evaluable. Assays for which no clear consensus was obtained or for which you were unable to obtain a clear result, as indicated by "I", were not included in either the assay or sample score calculation. At the bottom of each sample column on your result sheet you will find the number of assays performed by your lab for the sample, the number of results that were evaluable and used to calculate the score, and the number of 'correct' answers. The actual sample score as % 'correct' answers was calculated by dividing the number of 'correct' answers by the number of evaluable answers x 100. Finally, we also calculated an overall sample score as the average of the three individual sample scores. At this time we did not assign a grade, but may do so in the future. If any of your results are different from the corresponding consensus we ask that you take a careful look at your analysis and investigate why you may have reported a discrepant result. While this may be because of your assay's design and/or sensitivity and thus does not represent an error *per se,* it could also be a true error, indicating suboptimal performance of your assay, or be due to a contamination in case of apparently false positives.

NYS#L/L 2014-01 (Table 1)

B-cell tests: For IGH and IGK, there was unanimous agreement that these genes were rearranged, except for one lab that did not detect the IGK rearrangement. Rearrangements in IGH were detected with all primers irrespective of their provenance, except for Biomed-2 tube E that targets the DH7 region (Table 2). Rearrangements in IGK were, with one exception, only detected with tube A primers that target the Vk-Jk region (Table 3). No lab reported a fusion between IGH and BCL2; however, seven labs (100%) reported a fusion between IGH and CCND1. Ten of the twelve labs (83%) that tested for IGHV detected a mutation rate of <2% (range 1.0-1.4%) and thus did not call this sample hypermutated. The two other labs detected a mutation rate of 2.0% and 2.4%, respectively, and consequently called this sample hypermutated for IGHV. In conclusion, these results suggest that this sample contained a B-cell clone with IGH and IGK gene rearrangements and the IGH/CCND1 fusion gene.

T-cell tests: 13 out of 15 labs (87%) that tested for TRB by PCR found no rearrangement, and 21 out of 26 labs (81%) that tested for TRG reported no rearrangement (Table 1). However, one and four labs, respectively, detected a rearrangement in these genes. These labs should reexamine their results, and in particular verify that they don't have a PCR contamination. Together, these results suggest that this sample did not contain a clonal cell population with T-cell receptor gamma and/or beta gene rearrangements.

Other translocations: No translocations/fusions were detected at any of the loci tested, except for the IGH/CCND1 fusion mentioned above.

Various mutations (Table 8): No mutations were detected in any of the genes analyzed.

EBV: Five labs (100%) reported the presence of EBV DNA.

The results from all other tests performed were negative.

In aggregate, these results indicate that the sample contained a B-cell clone with IGH and IGK rearrangements and an IGH/CCND1 (Bcl-1) fusion gene, suggesting the presence of Mantle cell lymphoma cells. The results from Flow Cytometry indicated B cells with a plasmablast-like phenotype that exhibited surface expression of CD11b, CD19, CD20, and HLA-DR and expression of lambda and CD33, but that were negative for CD5 and CD10.

NYS#L/L 2014-02 (Table 1)
B-cell tests: For IGH and IGK, there was unanimous agreement that these genes were not rearranged except for one lab that reported an indeterminate result for IGH. Furthermore, no lab reported a fusion gene involving either the IGH/BCL2 or IGH/CCND1 loci. Thus, the overall conclusion is that this sample did not contain a clonal cell population with immunoglobulin gene rearrangements.

T-cell tests: 19 out of 26 labs (73%) that tested for TRG by PCR found a rearrangement that was detected with the Biomed-2 tube A primers, LDT primers targeting the Vγ1-8 region, and IVS v2.0 primers (Table 5). Fourteen out of fifteen labs (93%) that tested for TRB reported a rearrangement that was detected primarily with the Biomed-2 tube B primers (Table 4). Although the result for TRG is just short of the 80% needed for a consensus it is likely that this sample contained a clonal T-cell population with T-cell receptor beta and likely gamma gene rearrangements. Those labs that did not detect the TRG rearrangement are encouraged to reevaluate their result.

Other translocations: No translocations/fusions were detected.

Various mutations (Table 8): Five out of six labs (83%) detected the codon 61 mutation c.181C>A, p.Q61K, in N-ras. In addition, one mutation each was found in NOTCH1 and ASXL1.

In aggregate, these results indicate that the sample contained a clonal T-cell population with a N-ras mutation. The results from Flow Cytometry indicated an immature T-cell phenotype that expressed CD2, CD5, CD7, but was negative for surface CD3, CD4, TCR, CD10, CD11b, CD11c, and HLA-DR, and had dim expression of CD8.

NYS#L/L 2014-03 (Table 1)
B-cell tests: For IGH and IGK, there was unanimous agreement that these genes were rearranged including the one lab reporting oligoclonal for IGK. Rearrangements in IGH were detected with all primers irrespective of their provenance, except for Biomed-2 tubes D and E that target the DH1-6 and 7 regions, respectively (Table 2). Rearrangements in IGK too were detected with all primers (Table 3). No lab reported a fusion between IGH and BCL2 or CCND1, respectively. Mutation rates in IGHV ranged from 0-0.68% and thus this sample was unanimously classified as not hypermutated. In conclusion, these results suggest that this sample contained a B-cell clone with IGH and IGK gene rearrangements.

T-cell tests: 18 out of 25 labs (72%) that tested for TRG by PCR found a rearrangement that was detected by the Biomed-2 tube A primers, some LDT primers targeting the Vγ1-8 region, and the IVS v2.0 primers (Table 5). This result is just short of the 80% needed for a consensus. In contrast thirteen out of fifteen labs (87%) that tested for TRB reported no rearrangement (Table 4). Thus, this sample contained clonal cells with clear rearrangement of the immunoglobulin genes, and possibly TRG rearrangement.

BCR/ABL1: 30 labs detected a bcr/abl1 fusion product. Of these, 24 detected the p190 fusion product and five were not able to distinguish the p210/p190 fusion products. One lab reported the p210 fusion product, though we suspect that this is a data entry error. Most labs reported their result quantitatively, which ranged from 2.12% to 1323% relative to the respective housekeeping gene (Fig. 1 and Table 10), with a median of 61%. No other translocations/fusions were detected.

Various mutations (Table 8): Two labs detected the CALR variant c.1191_1199del9, which is an in frame deletion and thus more likely a polymorphism than a cancer mutation. A third lab also tested for CALR mutations, but did not report any mutation. Whether they did not detect this deletion, or whether they chose not to report it because it is likely benign is not known. No other mutations were detected in any of genes analyzed.

The results from all other tests performed were negative.

In aggregate, these results indicate that the sample contained a B-cell clone containing the bcr/abl1 p190 fusion gene and possibly a TRG rearrangement. The results from Flow Cytometry indicated prolymphocytic cells of B-cell lineage that expressed surface CD10, CD19, CD34, and HLA-DR, but that were negative for CD5, CD11b, CD11c, CD13, CD15, CD20, CD23 and CD33.

General comments

The attached tables show summaries of the results both overall (Table 1), as well as for each individual primer mix for the B- and T-cell tests (Tables 2-7). Furthermore, Table 8 shows a summary of the mutation results, and Table 9 shows summaries of the methods and reagents used for most of the tests. Table 10 and Figure 1 show the bcr/abl quantification distribution for sample L/L 2014-03. Figure 2 shows the DNA and RNA yield distributions for the three samples. Again the differences in yield are large ranging from 40- to 3337-fold, raising the question whether everybody reported their results the same way. Please make sure that you report the DNA and RNA yields in microgram (µg) and based on the actual volume of the original blood sample from which you isolated the DNA and RNA. Do not report the volume as the volume in which you eluted the nucleic acid into. We realize that shipping the samples at room temperature is suboptimal for subsequent RNA analysis. However, because of the combined shipping with the malignant immunophenotyping samples we cannot change that.

Finally, we would like to add some general comments. You really need to follow our instructions for filling out the result form, or we cannot guarantee correct evaluation of your results. **You must select the overall result in the first column**, as it is this result that is used in the evaluation. Then fill in or select the part of the additional information as appropriate. Also please make sure that you choose the correct method where there is a choice. If your starting material is DNA you must choose PCR. If your starting material is RNA, you must choose RT-PCR. Please note: RT stands for **R**everse **T**ranscription, not real time, and thus should only be used for assays whose starting material is RNA. Furthermore, we ask that if you obtain your primers/kits from InVivoScribe you correctly identify the source as IVS (not Biomed-2) (identified as gene rearrangement assays in their catalog) or IVS (Biomed-2) (identified as gene clonality assays in their catalog); for the purpose of this PT evaluation they are not considered lab developed even if you obtain the individual primer tubes separately as ASR reagents instead of as part of a RUO kit. This will make it easier to compare the performance of individual primer mixes. Finally, we ask that you analyze the samples by all molecular tests performed in your lab for which you hold or have applied for a NYS permit.

Some of you reported problems with submitting the result form from the 'Submit' button. Though we still are trying to figure out what the cause for this problem was as not everybody seemed to be affected, we strongly suspect it is connected to our mandated system wide migration to Windows 7 because of the Windows XP end-of-life. We apologize for the

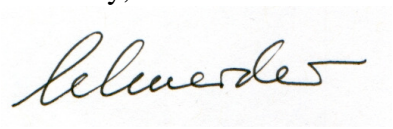
difficulties some of you had and will hopefully identify and rectify the problem before the next PT.

If you have any questions, comments or suggestions, you may contact me by phone or email at 518-474-2088 or schneid@wadsworth.org. For specific questions about your lab's report or the evaluation please contact Dr. Rong Yao at (518) 474-1744 or yaor@wadsworth.org, or Ms. Susanne McHale at (518) 486-5775 or smchale@wadsworth.org.

The next Molecular and Cellular Tumor Marker PT mail-out in 2014 will be:

Mail-out date	**Due Date**
October 28, 2014	November 26, 2014

Sincerely,



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New York State Molecular and Cellular Tumor Marker Proficiency Test Event MCTM 3-2014
Table 1: Summary of results

Assay / Sample	L/L 2014-01					L/L 2014-02					L/L 2014-03				
	R/H	G/U	I	O/N	Cons#	R/H	G/U	I	O/N	Cons#	R/H	G/U	I	O/N	Cons#
IGH	27				R		26	1		G	27				R
IGK	14	1			R		15			G	14			1	R
TRB	1	13	1		G	14	1			R	1	13	1		G
TRG	4	21	1		G	19	7			I	18	7			I
IGHV	2	10			U				13	N		13			U
	POS	NEG	I			POS	NEG	I			POS	NEG	I		
IGH/BCL2		12			NEG		12			NEG		12			NEG
IGH/CCND1	7				POS		7			NEG		7			NEG
IGH/MYC															
	MUT*	WT*	I			MUT*	WT*	I			MUT*	WT*	I		
JAK2 V617F		30			WT		30			WT		29			WT
JAK2 Exon 12		11			WT		11			WT		11			WT
MPL		13			WT		13			WT		13			WT
FLT3 ITD		9			WT		9			WT		9			WT
FLT3 TKD		9			WT		9			WT		9			WT
NPM1		17			WT		17			WT		17			WT
CEBPA		8			WT		8			WT		8			WT
IDH1		6			WT		6			WT		6			WT
IDH2		4			WT		4			WT		4			WT
KIT		10			WT		10			WT		10			WT
	POS	NEG	I			POS	NEG	I			POS	NEG	I		
BCR/ABL1 p210		26			NEG		26			NEG	1	25			NEG
BCR/ABL1 p190		25			NEG		25			NEG	24	1			POS
BCR/ABL1 p210/p190		5			NEG		5			NEG	5				POS
	MUT*	WT*	I			MUT*	WT*	I			MUT*	WT*	I		
ABL Kinase domain		2		5	I		1		6	I	1	5	1	1	WT
	POS	NEG	I			POS	NEG	I			POS	NEG	I		
PML/RARA		13			NEG		12	1		NEG		13			NEG
AML1/ETO		8			NEG		7	1		NEG		8			NEG
NPM1/ALK															
ETV6/RUNX1		3			NEG		2	1		I		3			NEG
CBFB/MYH11		4			NEG		3	1		NEG		4			NEG
TCF3/PBX1		1			I		1			I		1			I
MLL/AF4		2			I		2			I		2			I
	MUT*	WT*	I			MUT	WT	I			MUT*	WT*	I		
TP53		6	1		WT		6			WT		6			WT
KRAS		11			WT		11			WT		11			WT
NRAS		6			WT	5	1			MUT		6			WT
HRAS		4			WT		3			WT		4			WT
BRAF		14			WT		14			WT		14			WT
	POS	NEG	I			POS	NEG	I			POS	NEG	I		
EBV	5				POS		5			NEG		5			NEG
Interpretation:	B-cell prolymphocytic leukemia with IGH and IGK rearrangements and IGH/CCND1 translocation; consistent with Mantle cell lymphoma					Acute lymphoblastic leukemia with TRB and possible TRG rearrangements and NRAS mutation.					Acute lymphoblastic leukemia with IGH, IGK, and possible TRG rearrangements and bcr/abl p190 translocation.				
Comments															

R: rearranged/clonal band detected; G: germline/no clonal band detected; O: oligoclonal; For IGHV only: H: clonal band detected and hypermutated; U: clonal band detected, but not hypermutated; N: no clonal band detected.

MUT: mutated; WT: wild-type; N: no fusion product detected; NEG: neagtive or not detected; POS: positive or detected; I: indeterminate, a clear interpretation is not possible.

#Consensus based on ≥80% concordance; I if no consensus or <3 results

*For details of which exons/codons were analyzed see table 7.

Table 2: Summary for IGH primer mixes

	L/L 2014-01			L/L 2014-02			L/L 2014-03		
	R	**G**	**cons**	**R**	**G**	**cons**	**R**	**G**	**cons**
LDT FR 1	4	0	**R**	0	4	**G**	4	0	**R**
LDT FR 2	8	2	**R**	0	10	**G**	10	0	**R**
LDT FR 3	9	1	**R**	0	10	**G**	10	0	**R**
Biomed-2 Tube A	9	0	**R**	0	9	**G**	9	0	**R**
Biomed-2 Tube B	10	0	**R**	0	9	**G**	10	0	**R**
Biomed-2 Tube C	10	0	**R**	0	10	**G**	10	0	**R**
Biomed-2 Tube D	2	2	**I**	0	4	**G**	1	3	**I**
Biomed-2 Tube E	0	5	**G**	0	5	**G**	0	5	**G**
IVS FR 1	6	0	**R**	0	6	**G**	5	0	**R**
IVS FR 2	7	0	**R**	0	7	**G**	7	0	**R**
IVS FR 3	8	0	**R**	0	8	**G**	8	0	**R**

Table 3: Summary for IGK primer mixes

	L/L 2014-01			L/L 2014-02			L/L 2014-03		
	R	**G**	**cons**	**R**	**G**	**cons**	**R**	**G**	**cons**
LDT Tube A	7	1	**R**	0	8	**G**	7	0	**R**
LDT Tube B	1	7	**G**	0	8	**G**	7	0	**R**
Biomed-2 Tube A	8	0	**R**	0	8	**G**	8	0	**R**
Biomed-2 Tube B	0	8	**G**	0	8	**G**	8	0	**R**

Table 4: Summary for TRB primer mixes

	L/L 2014-01			L/L 2014-02			L/L 2014-03		
	R	**G**	**cons**	**R**	**G**	**cons**	**R**	**G**	**cons**
LDT Tube A	0	3	**G**	1	2	**I**	0	3	**G**
LDT Tube B	0	3	**G**	2	1	**I**	0	3	**G**
Biomed-2 Tube A	0	10	**G**	2	9	**G**	0	11	**G**
Biomed-2 Tube B	1	9	**G**	11	0	**R**	0	11	**G**
Biomed-2 Tube C	0	8	**G**	0	10	**G**	1	8	**G**

Table 5: Summary for TRG primer mixes

	L/L 2014-01			L/L 2014-02			L/L 2014-03		
	R	**G**	**cons**	**R**	**G**	**cons**	**R**	**G**	**cons**
LDT Vγ1-8	1	7	**G**	3	5	**I**	3	5	**I**
LDT Vγ9	2	5	**I**	0	7	**G**	0	7	**G**
LDT Vγ10	1	4	**G**	0	5	**G**	0	5	**G**
LDT Vγ11	0	4	**G**	0	4	**G**	0	4	**G**
Biomed-2 Tube A	2	7	**I**	9	1	**R**	9	1	**R**
Biomed-2 Tube B	2	7	**I**	0	10	**G**	0	10	**G**
IVS Mix 1	0	2	**I**	1	1	**I**	1	1	**I**
IVS Mix 2	0	2	**I**	0	2	**I**	0	2	**I**
IVS v2.0	0	4	**G**	4	0	**R**	4	0	**R**

Table 6: Summary for BCL2 primer mixes

	L/L 2014-01			L/L 2014-02			L/L 2014-03		
	POS	**NEG**	**cons**	**POS**	**NEG**	**cons**	**POS**	**NED**	**cons**
LDT MBR	0	3	**G**	0	3	**G**	0	3	**G**
LDT MBR3'	0	0		0	0		0	0	
LDT mcr	0	2	**I**	0	2	**I**	0	2	**I**
Biomed-2 Tube A	0	2	**I**	0	2	**I**	0	2	**I**
Biomed-2 Tube B	0	2	**I**	0	2	**I**	0	2	**I**
Biomed-2 Tube C	0	2	**I**	0	2	**I**	0	2	**I**
IVS Mix1b	0	0		0	0		0	0	
IVS Mix2b	0	0		0	0		0	0	

Table 7: Summary for PML/RARA primer mixes

	L/L 2014-01			L/L 2014-02			L/L 2014-03		
	POS	**NEG**	**cons**	**POS**	**NEG**	**cons**	**POS**	**NED**	**cons**
Long	0	8	**G**	0	8	**G**	0	8	**G**
Short	0	8	**G**	0	8	**G**	0	8	**G**
Varaible	0	2	**I**	0	2	**I**	0	2	**I**
L/S/V not distinguished	0	1	**I**	0	1	**I**	0	1	**I**

Table 8: Summary of mutation assay results including polymorphisms

		L/L 2014-01		L/L 2014-02		L/L 2014-03	
Gene	**exons/codons tested**	**Result (WT if not indicated)**	**# of variants detected**	**Result (WT if not indicated)**	**# of variants detected**	**Result (WT if not indicated)**	**# of variants detected**
JAK2 Exon 12							
JAK2 Exon 13							
JAK2 exon 14	codon 617						
MPL	codon 515						
	codon 505/515						
	amino acid 490 to 520 region						
	W515L/K						
	S505, W515						
	exon 10						
FLT3 TKD	D835						
	Exon 20						
CEBPA	Entire coding region, 1 exon.						
	exon 1						
	exon 1						
	all coding						
	chromosome 19, single exon						
IDH1							
IDH2							
KIT	exon 17						
	Exons 8, 9, 11, 13, 17						
	exon 8/17						
	D816; exons 8 & 17						
TP53	exon 5-9						
	Exons 4, 5, 6, 7, 8, 9						
	exon 2-11						
KRAS	codon 12/13/61						
	exon 1, 2						
	codon 12/13						
NRAS	codons 12/13/61			Q61K	3		
	exon 1,2			p.Q61K, c.181C>A, HETEROZYGOUS	1		
	exons 2-3			c.181C>A (p.Q61K)	1		
HRAS	codons 12/13/61						
	exon 1,2						
BRAF	codon 599-602, exon 15						
	codon 600						
	exon 11, 12, 15						
	Exons 11,12,15, codon V600						
	V600E ONLY						
EGFR	exon 19/858						
	Exon 19 del						
	L858						
PIK3CA	Exons 1,9,20						
PDGFRA	Exons 12, 18						
WT1	exons 7 & 9						
	exons 7 & 9						
MYD88	codon 265						
NOTCH1	exon 34			c.7544_7545delCT; p.P2515fs.	1		
ASXL1	Ex13 (Some refer it as Ex12)			c.1934delG (p.G645fs); c.2019C>T (p.G673G)	1		
CALR	Exon 9					c.1191_1199del9; p.D397_D400delinsD	1
	Exon 9					c.1191_1199del, p.Glu398_Asp400del	1
RUNX1	exon 1-8						

NOTE **For each gene the area analyzed is listed with the number of labs reporting variants.**
No entry in the result columns means no specific mutation data were reported.

Table 9: Summary of methods and reagents used

	Total	SB	PCR	Seq	PCR + Seq	NGS	Lab developed	IVS (Biomed-2)	IVS (not Biomed-2)	IVS TRG 2.0	IVS Lymphotrack	Lab developed and IVS (Biomed-2)	Lab developed and IVS (not Biomed-2)	Qualitative	Quantitative	0
IGH	28	0	28	0	0	0	9	10	8	0	0	0	0	0	0	0
IGK	16	0	16	0	0	0	3	11	0	0	0	0	0	0	0	0
TRB	15	0	15	0	0	0	2	12	0	0	0	0	0	0	0	0
TRG	27	0	27	0	0	0	13	9	1	3	0	0	0	0	0	0
IGHV	13	0	2	7	4	0	5	0	0	0	0	0	0	0	0	0
IGH/BCL2	12	0	12	0	0	0	4	1	0	0	0	0	0	10	1	0
IGH/CCND1	7	0	7	0	0	0	1	0	0	0	0	0	0	5	2	0
IGH/MYC	0	0	0	0	0	0	0	0	0	0	0	0	0			0

	Total	PCR	RT-PCR	Seq	PCR + Seq	RT-PCR + Seq	NGS	Lab developed	Ipsogen (Qiagen)	IVS	Qualitative	Quantitative	Qual and Quant
JAK2 V617F	30	24	0	2	1	2	1	23	7	0	18	7	5
JAK2 Exon 12	11	4	0	3	1	1	2	10	0	0			
MPL	13	4	0	5	0	2	2	12	0	0			
FLT3 ITD	8	8	0	0	0	0	0	8	0	0			
FLT3 TKD	9	6	0	1	0	0	2	7	0	0			
NPM1	17	15	0	0	0	0	2	15	0	0			
CEBPA	8	2	0	5	1	0	0	8	0	0			
IDH1	6	1	0	3	0	0	2	4	0	0			
IDH2	4	0	0	3	0	0	1	3	0	0			
KIT	10	3	0	2	3	0	2	8	0	0			

	Total	PCR	RT-PCR	Seq	PCR + Seq	RT-PCR Seq	NGS	Lab developed	Ipsogen (Qiagen)	Roche	Cepheid	Asuragen	Qualitative	Quantitative	Qual and Quant	IS Normalized
BCR/ABL1 p210	26	0	26	0	0	0	0	19	6	0	1	1	3	19	4	14
BCR/ABL1 p190	25	2	23	0	0	0	0	18	5	0	0	1	6	15	4	
BCR/ABL1 p210/p190	5	0	5	0	0	0	0	3	0	2	0	1	1	5	0	
Abl Kinase domain	9	0	2	2	1	3	1	8	0	0	0	0				
PML/RARA	13	2	11	0	0	0	0	13	0	0	0	0	4	8	1	
AML1/ETO	8	2	6	0	0	0	0	7	0	0	0	0	4	4	0	
NPM1/ALK	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
ETV6/RUNX1	3	0	3	0	0	0	0	3	0	0	0	0	1	2	0	
CBFB/MYH11	4	0	4	0	0	0	0	4	0	0	0	0	2	2	0	
TCF3/PBX1	1	0	1	0	0	0	0	1	0	0	0	0	1	0	0	
MLL/AF4	2	0	2	0	0	0	0	2	0	0	0	0	0	0	0	

	Total	PCR	Seq	PCR + Seq	NGS	Lab developed	Qiagen	Roche Cobas	Assuragen/ Luminex	Sequenom/ Massspec	Other
TP53	7	0	5	1	1	6					1
KRAS	11	1	4	3	1	6	2		2		
NRAS	6	0	3	2	1	4	1				1
HRAS	4	1	1	1	1	3					1
BRAF	14	7	4	2	1	6	3			1	2
EBV	5	3	0	0	0	3					

NOTE: any discrepancies between the numbers in this table and the number of results in Table 1 are caused by incomplete and/or inconsistent data submission by some labs

Figure 1: L/L 2014-03 bcr/abl quantification

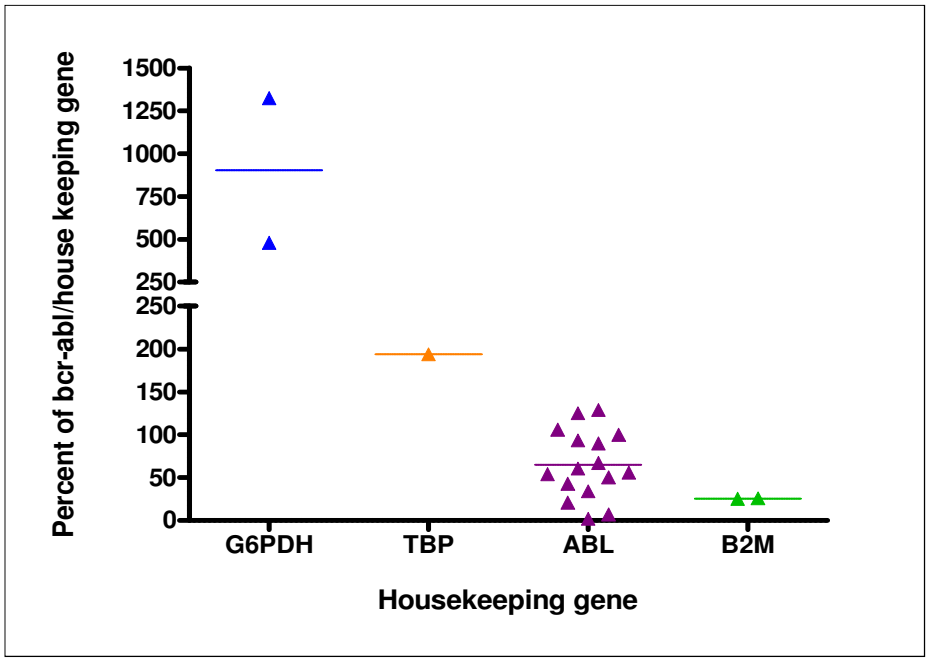
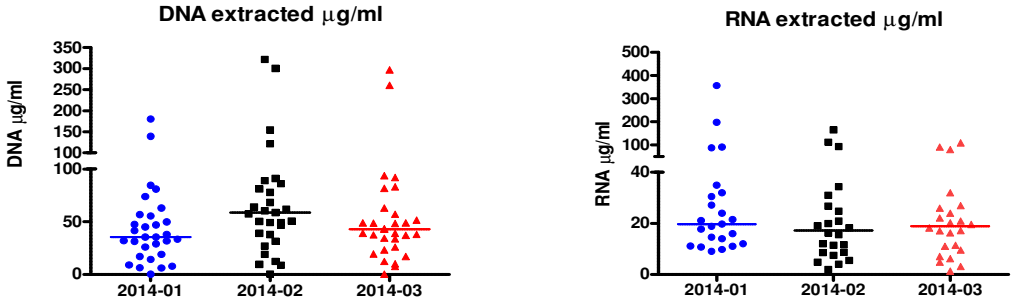


Table 10: Results of bcr/abl qRT-PCR assays for NYS MCTM L/L 2014-03

	bcr-abl/G6PDH		bcr-abl/TBP		bcr-abl/BCR		bcr-abl/abl		bcr-abl/GUSB		bcr/abl/B2M	
	p210	p190	p210	p190	p210	p190	p210	p190	p210	p190	p210	p190
Median		902%		194%			111%	59%				26%
Min		480%		194%			111%	2.1%				25%
Max		1323%		194%			111%	129%				26%
N		2		1			1	16				2
Mean		902%		194%			111%	65%				26%
SD		596%						39%				0.3%

Figure 2: NYS MCTM PT 03-2014 DNA and RNA yields.
The yields were converted to ug DNA and RNA per 1 ml blood.



	L/L 2014-01	L/L 2014-02	L/L 2014-03		L/L 2014-01	L/L 2014-02	L/L 2014-03
	DNA	DNA	DNA		RNA	RNA	RNA
Mean	44.6	120.9	57.9	Mean	47.3	29.2	25.9
Median	35.5	58.1	41.0	Median	19.65	16.16	18.17
Min	0.28	0.45	0.3	Min	9.0	1.9	1.3
Max*	180.0	1502.0	296.8	Max	356.7	165	109

*Graph excludes the max DNA yield for L/L 2014-02 because it was far out of line with all other results.