

Appendix C: Itemization of Focus Group Comments

Topic	Comment	No. of comments	Notes
Sample Acceptance/ Rejection Criteria	Samples are currently opened in a timely fashion and recorded efficiently	2	
	Criteria for acceptable sample volumes	2	Can EDTA:blood influence quality (ie when 1 ml blood is received in a 10 ml EDTA tube)
	Clearer definitions of irretrievable samples/exceptional circumstances in which samples may be processed despite not meeting acceptance criteria		
	Review acceptable sample transport times	2	
	Establish clear acceptance/rejection criteria with strict adherence to criteria (except in cases of irretrievable samples)	11	ie.samples < 5 days old, EDTA collection tube, not clotted, not hemolyzed, completed req
	Establish protocol for documentation and follow-up of sample rejection		
	Establish reporting system for rejected or sub-optimal samples	2	
	Education to external sites re: proper conditions	3	development of a reject report and information pamphlet regarding appropriate sample conditions
	Generation of a log for tracking receipt of non-ideal samples.		
	Improve communication with external laboratories that regularly send samples		
	Establish a policy on receiving samples on Fridays/ before long weekends		
	Consider storing bloods at 4°C until ready to process		
	System of quick labels are great; helps to get process going		
	Prioritize sample processing		ie. based on days since blood draw, type of test (with those requiring high molec weight being a higher priority)
	Address completion of reqs (to avoid delays in sample processing, ie, if test required is not clear)	2	
	Develop a better system for recording/tracking conditions of compromised/ sub-optimal samples		
	Improve record keeping of communications with physician's office/ counsellors regarding incomplete requisitions or concerns related to the sample/test requested		
DNA Quality Indicators			
	sample conditions at receipt should be recorded	6	ie. clearly record sample conditions (hot, frozen et...) on req and in comments on MLAB
	mark DNA aliquots if there were poor sample conditions		ie. use a red do of label/tube to mark potentially poor samples
	record time from blood draw to extraction for each sample (blood age)	5	
	maintain OD readings	6	
	record OD readings in MLAB		
	maintain use of integrity gels	7	
	Establish protocol to ensure consistency in gel/ electrophoresis conditions for integrity gels		
	Examine factors that influence reproducibility of OD readings of a given sample		
	correlation of successful testing with DNA sample		
	log lot #s of all reagents used in extractions		
	examine dissolved DNA (more low TE needed)	2	inconsistency in DNA reconstitution impacts ability to take accurate OD readings/integrity gels
	record heating block temperature		
	record #s of sample received daily/#s extracted by each method		

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	record whether sample has been split or extracted from a separate sample		
	record any irregularities during extraction	2	ie. time delays, odd pellet, jelly-like pellet, brownish pellet, etc.
	record results of integrity gels on data pending sheets and in MLAB		
DNA Extraction Protocol	Use small aliquots of DNA extraction solutions and discard after extraction	3	
	review critical steps of the procedure and indicate these steps in written protocol	2	ie. indicate in protocol where there is flexibility/no flexibility including incubation times
	establish cut-off times for samples to be included in an extraction batch		
	examine hydration process and establish procedures for ensuring better consistency in how well DNA is dissolved		
	minimize/alleviate deviations from manufacturer's protocol	3	do any current deviations impact DNA quality?
	ensure consistency in extraction from either whole blood or buffy coat	2	
	test new lot #s prior to use	2	
	establish consistency in the extraction process	5	ie. not having rotating techs; consistency in timing, techniques, etc.), potential use of robots
	establish checklist fo keeping track of any variables during extraction	2	"more stringent testing if extreme variation has been noted"
	record lot #s used in extraction		
	limit batch sizes	3	
	evaluate possibility of using different extraction kits		
	evaluate quality of reagents that may affect DNA sensitivity at specific stages		
	need a better method for aliquoting/using reagents		
	examine using a vortexer with a tube holder		
	extraction from beginning to end in 1 day		
	avoid making notes on protocols; change in Paradigm if applicable		
	follow through from start to finish without delays		
	avoid multitasking when doing extractions		
	POUR 3ml whole blood in 15 ml tubes then add 9ml RBL to ensure that all samples are lysed for the same amount of time		
Extraction Supplies and Reagents	store 70% EtOH and 100% isopropanol in brown, light-protected bottles		
	examine storage and aliquot conditions of alcohols to ensure % stability	2	use squeeze bottles(?), aliquot into 15 ml tubes
	ensure appropriate size of aliquots for single day use	3	
	examine whether there are specific requirements for plastic-ware		does plastic-ware require autoclaving
	must use HIGHEST quality/most appropriate reagents available for molecular techniques	3	
	test new lot #s prior to putting into use	2	
	purchase a new extractor that will provide sufficient DNA for ALL tests		
	purchase higher grade ethanol and isopropanol	2	
	ensure temperature of heating block can be measured accurately		

	avoid "topping up" solutions: discard remaining reagents and open new lot #s		"a little bit of waste is better than guessing"
Topic	Comment	No. of comments	Notes
	dedicate ordering		
	change storage of A.P. kits		
	Record lot #s used in extraction	4	
	Record date reagents opened	2	
	Individual aliquots should be discarded after 2 days		
	Poll other labs to see what kits they are using		
	Keep tubes, tips, lids, closed in hood	2	
	Ensure serility and sterile techniques		
	confirm that reagents not past expiry dates	2	
	examine whether all reagents are being stored in proper conditions		
	purchase robot for extraction		
	assay problems almost always come back to water		
Notification of Testing Failures at External Labs Using DNA from CHEO MGDxL	Include something in package for all send-outs to request info regarding DNA quality/ability to complete test requested using DNA provided	5	formats for requests could include forms, postcards, questionnaires, email address for providing comments re: DNA quality to
	Request report from all counsellors/physicians re: success/failure in Send-Out report to physician		
	Run integrity gels more than one on certain samples after storing		
	simplify send-out process		
	ship whole blood whenever possible		
	Provide education for providers regarding importance of notifying Laboratories of issues when they arise		
DNA storage	Run integrity gels more than once on certain samples during storage to monitor quality over time		
	Examine appropriate temperature for long term storage	5	ie. 4, -20, or -80 degrees
	DNA sitting on bench for extended period of time		
	Discard old DNA samples in a timely manner		
	Look at state of the art on storage		
	Encourage the formation of a provincial DNA bank		ie. we are not a banking facility
	Only store DNA for 3 months then hand over to physician or discard		
	Establish a better method of keeping track of what is being stored and what has been discarded		
	Lyophilize all DNA		