



Original Sample QC

General Information

Order Number	HN00117803	Name of Customer	Nivest Prayoonthien	Date of Order	2019-11-07
--------------	------------	------------------	---------------------	---------------	------------

Final QC Result of DNA sample(s)					
Arrival Date	Experiment Date	Sample count	Pass	Fail	Hold
2019-11-08	2019-11-11	4	4	0	0

Final QC Result of RNA sample(s)					
Arrival Date	Experiment Date	Sample count	Pass	Fail	Hold
N/A	N/A	N/A	N/A	N/A	N/A

The QC criteria refer to the specification requirements of a single run. In any cases, we may encounter the shortage of sample volume or amount due to various reasons such as a library construction failure. In these cases a request of an additional sample will be inevitable.

Therefore, we recommend double the amount to be supplied at first place to minimize any delay of the whole procedure.

*** Pass :** Proceed with the library construction.

*** Fail :** Further processes are on hold until the replacement samples received.

We do not recommend in proceeding further steps until a specific instruction was given from the client.

*** Hold :** A specific instruction should be given by the client for further processing as the QC pattern may be triggered by the sample nature.

MacroGen does not proceed the next step until we have received your permission.

As 3 ul was taken from the sample for sample (library) QC purposes, the indicated volume represents 3ul less than the total volume received.

QC Result of DNA

Arrival Date	2019-11-08	Experiment Date	2019-11-11	Tested by	CJH
Comment					

#	Sample Name	Conc. (ng/ul)	Final Volume (ul)	Total Amount (ug)	Result*	
1	Vermicompost	14.544	35	0.509	Pass	
2	M_pegunagut	0.263	35	0.009	Pass	
3	M_posthrumagut	0.233	35	0.008	Pass	
4	Perionyx_sp1gut	1.807	35	0.063	Pass	

DNA QC Method

1. Quantity of DNA : Done by picogreen* method using Victor 3 fluorometry.

Macrogreen quantifies the starting genomic material by a fluorescence-based quantification, rather than a UV-spectrometer-based method.

This is because fluorescence-based methods, which employ a double-stranded DNA specific dye, will specifically and accurately quantitate dsDNA even in the presence of many common contaminants. UV spectrometer methods based on 260 OD readings are prone to overestimating the DNA concentration due to the presence of RNA and other contaminants commonly found in gDNA preparations.

* Picogreen (Invitrogen, cat.#P7589)

2. Assessing the condition of the DNA : Done by gel electrophoresis method.

Gel electrophoresis is a powerful means for revealing the condition (including the presence or absence) of DNA in a sample.

Impurities, such as detergents or proteins, can be revealed by smearing of DNA bands. RNA, which interferes with 260 nm readings, is often visible at the bottom of a gel.

A ladder or smear below a band of interest may indicate nicking or other damage to DNA.

3. Size Check of DNA [Optional , Upon request, Charged]

1) DNA fragments <1kb : 2100 Bioanalyzer* is used for checking the size.

*Macrogreen use DNA 1000 chip and DNA 7500chip for normal PCR product, high sensitivity chip for very small amount of DNA fragment such as ChIPed DNA.

*<http://www.genomics.agilent.com/CollectionSubpage.aspx?PageType=Product&SubPageType=ProductData&PageID=1636>

2) DNA fragments < 150kb : PFGE method is used for large size of DNA fragment. Please keep in mind that we conduct this upon request and will be charged.

RNA QC Method

1. Quality & Quantity Check of RNA : 2100 Bioanalyzer*** (or 2200 TapeStation****) is used.

We check total RNA integrity using an Agilent Technologies 2100 Bioanalyzer (or 2200 TapeStation) with an RNA Integrity Number (RIN)*** value greater than or equal to 7.

RNA that has DNA contamination will result in an underestimation of the amount of RNA used. We recommend including a DNase step with the RNA isolation method.

However, contaminant DNA will be removed during mRNA purification. It is very important to use high-quality RNA as the starting material.

Use of degraded RNA can result in low yield, over-representation of the 5' ends of the RNA molecules, or failure of the protocol.

***<http://www.genomics.agilent.com/CollectionSubpage.aspx?PageType=Product&SubPageType=ProductData&PageID=1648>

****<http://www.genomics.agilent.com/article.jsp?crumbAction=push&pageId=900109>

DNA QC Criteria

Platform	Library Type	Library Kit	Total Amount	DIN	etc
MiSeq	Amplicon DNA library	Metagenome Amplicon	-	-	Conc > 0.1ng/ul
MiSeq	Amplicon DNA library	target amplicon DNA	-	-	Conc > 0.1ng/ul