Molecular Research LP

MR DNA

Shipping Instructions Molecular Research DNA Lab (MR.DNA)

Shipping from Outside the US to MRDNAlab.com :

Please notice our address

Molecular Research Laboratory
503 Clovis Road
Shallowater, TX 79363
United States
806-789-7984
Contact: Dr. Scot E. Dowd

http://www.mrdnalab.com

Molecular Research DNA-RNA LAB highly recommends that customers ship their valuable specimens using FEDEX

The following are "full service" carriers.

FedEx http://fedex.com
DHL http://www.dhl.com
UPS http://www.ups.com

***Please note**: In our experience using any company other than FedEx and not following the instructions here precisely may cause your purified DNA samples to be held up in customs. There are many advantages to using a full service carrier. DHL and UPS are also good reliable carriers.*

With any shipper please choose door to door service.

IMPORTANT NOTE: Control samples .. MR DNA does not automatically run control samples for your study. We highly recommend that you provide DNA extraction controls, blank samples, mock communities etc. needed for your study .. if you do not know what to include for these, we are happy to provide more detailed advice of course.

1. **If we are extracting DNA for your study, we are happy to extract a blank sample and include it in your study to act as a DNA extraction control for background $25 per control extraction**
2. **We have inhouse mock communities we can use along with your study to act as a positive controls simply request this service.**
3. **If your samples are sent in a storage media we recommend sending us a sample of the storage media without sample added to use as a separate extraction control.**

Almost without exception any reagent used in a laboratory can have some background signal especially for 16s .Please take into consideration when designing your study.

1. **If you are sending DNA we recommend to extract a blank and send that along with your samples (PLEASE label it as an extraction blank so we do not try to optimize it or make it work like a high biomass sample).. blank extractions usually generate a very weak signal and we run them just like a normal sample to give your study a more accurate background**
2. **Sending a mock community .. standard mock communities are available from ATCC for instance, but we do have these available inhouse and we can include them in your study on request.**

We do charge for all control samples we run just like a normal sample so please let us know about the controls and again do not blind the naming of controls as we do not want to spend time trying to figure out why a blank is not generating a strong signal like a normal sample (this costs us extra time and money and it will cost you extra time and money) .. we want to treat the controls like normal samples to provide the best possible background or mock community data for your study.

DNA/RNA: HOW MUCH DNA DO I SEND?

DNA What are specifications for DNA :

1. **DIVERSITY AND AMPLICON such as 16s:**
   NOTE We do not have a concentration requirement. Send 20-30ul of the best DNA you can and we will do the best we can. We can work often with very low concentrations of DNA such as 1ng/ul without issue. the most important aspect is good pure DNA without inhibitors. But as noted above send 20ul of your DNA as fresh and pure as possible and we will do our best to work with it. **With LOW DNA control samples are very important please see previous notes on control samples!!!**

   With amplicon sequencing such as 16s or similar we need enough DNA to create several PCR reactions and to allow for optimization (getting all the samples to amplify uniformly as much as possible). Send 20ul and the best concentrations possible (Best to send normalized concentrations across all samples if you deem this necessary. We can provide normalization services upon request. A good

nominal concentration is 20ng/ul. However, we do not have a strict requirement and the only thing we require is "Send the best DNA you can… the better the DNA in all samples the better the results and faster we can deliver your data to you".

2. **GENOME SEQUENCING:**
   a. **ILLUMINA GENOME AND METAGENOME: With genome or metagenome we need at approximately 500ng-1ug total high molecular weight GOOD DNA in 20-30ul. It is much better to send at least 2ug of DNA to allow us to optimize the sequencing libraries. Send the best concentration you can e.g. 30ng/ul is reasonable. We can work with less DNA also and even very low amounts of DNA with some additional services that are available such as whole genome amplification or linear amplification etc.**
   b. **PAC BIO** For best results we need up to 2ug of purified high molecular weight DNA .. as above we can work with a little less DNA .. if much less we do have low input methods $100/sample to work with most sources of DNA that do not achieve the standard amount noted above

WHAT IS GOOD DNA ?

the best data output and best sequencing results depends entirely on the quality of the DNA provided

DNA is best quantified using fluorescent methods such as qubit.. integrity of DNA may be best determined using gels.. contaminations best determined with spectrophotometray e.g. nanodrop

For genomics purified high molecular weight DNA (gentle extractions with slow pipetting and VERY gentle vortexing if needed) hard to do with some samples so please do as best you can .. molecular weight can be determined with a gel . for genomics most fragments > 10K are best .. larger if possible. If your organism has plasmids a separate plasmid extraction might be necessary as some genomic extraction do not help to isolate small plasmids sometimes .

Good-quality DNA will have an A260/A280 ratio of 1.7–2.0. A reading of 1.6 does not render the DNA unsuitable for any application, but lower ratios indicate more contaminants are present.

As a guideline, the A260/A230 is best if greater than 1.5.

RNA: Shipping RNA to maintain integrity (RNAstable reagent is great to allow shipping of purified RNA and maintaining its integrity

The requirements for RNA are variable.. Again send as much RNA of the best quality you can. With most RNAseq we will need 1-2ug of good RNA. 1ug may suffice for polyA selection and total RNA sequencing. Up to 2ug of RNA may be needed for metatranscriptomes with ribosomal depletion or for bacterial RNAseq with ribosomal depletion. The better the RNA quality the better the results. MR DNA will do the best we can with the RNA you send but if you want specific requirements during QC let us know (e.g. if you indicate that the RIN should be > 7 to proceed we will follow these requirements. Please note that subsequent QC for samples that need to be resent may incur additional costs. QC of samples is not free and the QC included in our base costs per sample include ONE QC per sample.

DO NOT SEND hidden or blinded controls AT ALL . We often do several rounds of troubleshooting and this is an undue burden on our time and resources when we run your negative controls over and over to try and get them to work. If you send a control label it as a positive or negative control (as the case may be). We will treat the control sample strictly as labeled or requested. Please note that positive controls must be handled separately from actual samples. Low diversity positive controls or samples with high concentrations or abundant DNA should not be sequenced with high diversity low input DNA samples. Let us know this we can separate low biomass from high biomass usually without additional cost. Several known factors related to massive parallel sequencing such as barcode drift will occur if samples of very different projects are run together. It is highly recommended that if you are sending multiple projects in the same shipment to include different submission forms for each project so that they can automatically be run separately. This can avoid too much problem with barcode shift and drift. Many investigators will send small projects batched together to save a little money. MR DNA is not responsible for factors related to this and strongly urge that projects that are shipped together be sequenced individually regardless. Very easy feel free to ask questions to clarify if this is not clear. We are here only to help!

New service: Slowing Barcode drift: Adds $15/sample. When synthesizing reagents there can and usually occurs some errors and some low level carryover from one barcode/primer to the next. This is not the fault of the primer companies ;-) . This effect is not usually large but with the scale of next generation sequencing it becomes noticeable especially when lo w biomass samples are run with large spike controls or high biomass samples (see above). We now have improved assays that can reduce this effect even further than normal HPLC primer purification. We have added this method to some of our more popular assays and will continue to add new assays as interest from the community increases. For now there is a modest upcharge of $15/assay for this.. we currently have this for assays 27F, 515F, and ITS1-2. Please inquire about this new service.

LOW BIOMASS: This mainly applies to genome, metagenome, transcriptome and metatranscriptomes sequencing. Not always relevant with amplicon sequencing. With any laboratory low biomass samples can present special challenges. MR DNA is a highly separated workflow facility. All of our processes are split into separately controlled sections and units. This design means that contamination potential is very low. With low biomass samples the best way to generate reliable results is to prevent contamination before and after samples arrive at MR DNA. The personnel at MR DNA are specifically trained to handle low biomass samples. MR DNA does have a specialized process for analyzing low biomass samples. This process adds $50/sample to any sequencing price and may ensure that your project obtains data faster and more reliably. Please notify or select the low biomass process for your samples that may require this treatment.

PACKAGING Suggestions for non-DNA/RNA samples. Samples for which MR DNA will provide nucleic acid extractions

There are no testing requirements for containers used to ship non-pathogenic cultures or non-infectious biological materials. **Recommended Packaging for Non-Pathogenic Biological Materials . If you are sending pathogens please consult with a scientists at MR DNA LAB.**

The main goal is the pack your samples securely to prevent leakage and contamination of other samples if your sample should leak. TO protect from breaking, to keep the lids from coming off, to prevent the samples from being damaged if the package is handled

roughly during shipment etc. If you are sending plates be absolutely sure that the covers will not lift off during shipping. Films, foils etc. during the pressure changes of shipping can lift off, pop off. . Even if a small portion of the film or foil lift. this will allow for cross contamination of your DNA, loss of samples etc. MR DNA is not responsible for anything related to issues that occur during shipping.

Primary container (the inner container holding the DNA tubes):

1. FILL OUT A Sample submission form (obtained from MR DNA via email)
2. Use a vial, tube or plate made of high grade plastic or other medium suitable for transportation of the material being shipped. 1.5 ml snaptop or 2ml screwtop are appropriate.
3. Seal each vial tube or plate securely (tape parafilm etc) to absolutely prevent leakage and breakage and evaporation
4. Clearly identify the contents and avoid abbreviations (e.g. write out Salmonella enterica MK-06 rather than just MK-06).
5. Place them inside the primary container so they are protected from being crushed and if a tube breaks be sure to include enough absorbent material (e.g. paper towels) to fully contain any liquid if a tube breaks. Paper towels and other absorbent material also provide protection from breaking.

Secondary container:

1. Use a watertight/leak proof container and reinforce with an adhesive tape as necessary to contain the individual vials contents (e.g. zip-lock type bag).
2. Include a packing list in a separate (zip lock bag or other water tight package) to prevent it from becoming wet and unreadable with a complete list of the contents including the scientific name and the amount in ml for liquids. Also email this list to MR.DNA. info@mrdnalab.com
3. Surround each primary container (e.g. vial) with sufficient absorbent packing material to completely absorb the contents should the primary container break Shipping container:
4. Use an outer container (shipping box) made of sufficient strength to protect the specimen.
5. Affix or write with permanent sharpie pen or other marking method a proper label on each tube or vial containing a sample to identify the contents and relate them to the packing list. Make sure labels cannot be wiped off.
6. Include on the packing slip an accurate address label with the complete address and phone number AND NAME for both the shipper and the recipient.
7. Do not list your secretary list the investigator or scientist so that the lab and MRDNA office personnel can get you your final data analyses without delays.
8. Affix the "double up arrows" sticker if orientation is important.
9. **using dry ice:** If dry ice must be used in the packaging, these materials must be declared, and packages must be properly packaged and labeled with an ORM class 9 label and a **Shipper's Declaration for Dangerous Goods must be completed (contact your shipper or shipping professionals at your institution for help with this)**. Note: Dry ice should never be placed in a sealed container. **MOST DNA samples are stable at room temperature for a few days and will not require dry ice.** If you do have any concerns please use your best judgment or contact MR DNA at info@mrdnalab.com for further instructions.

RNA: RNA is much less stable than DNA. Some suggestions for shipping RNA include lyophilization, shipping on dry ice, using RNAstable reagent, etc. For samples (which MR DNA

will extract RNA), ship these in an appropriate fashion to maintain the RNA profile. This can include shipping on dry ice or in RNAlater reagents (Qiagen). If you are unfamiliar with RNA handling procedures or RNA extraction procedures please consult with us and we will try to help you achieve a successful RNA experiment. Always send as much RNA or mRNA as you can!

Ship materials to:
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Contact: Dr. Scot E. Dowd