

DNA Sequencing



Oct 27, 2020

Dear Researchers,

RIOH (formerly MICB) has offered a DNA sequencing service since 1998 to members of the Institute as well as University researchers. Our initial service was offered using a single column ABI 310 sequencer; however, in May 2010, we entered an agreement with Eurofins MWG / Operon to provide this service for us at a preferred price. In the past weeks, pricing has changed to reflect the weaker Canadian dollar. Our current pricing is as follows:

1. **Single tube samples..... \$5.50 per sample**
2. **Single tube PowerRead samples \$8.50 per sample**
3. **Plate of 24-94* samples \$3.75 per sample**

Above costs includes: free shipping, [free in-house primers](https://www.eurofinsgenomics.com/en/orderpages/oligos/standard-primers/) (see <https://www.eurofinsgenomics.com/en/orderpages/oligos/standard-primers/>), free sample processing, free consultation and taxes. Where applicable, expect 800-1100bp of usable sequence.

4. **ShortSeq (800bp) plate of 94* samples with your primers added \$250 per plate (\$2.66 per sample)**
Cost includes: free shipping, free sample processing, free consultation. Samples must be provided with premixed primers in appropriate skirted PCR plate Fisherbrand Cat#14230237 with corresponding 8 strip flat caps Fisherbrand Cat# 14230230. Where applicable, expect 800bp of usable sequence.

Samples can be plasmids or PCR DNA.

**Wells G12 & H12 on plates reserved for in-house controls.*

Preferred plate Fisherbrand Cat# 14230237 with caps Fisherbrand Cat# 14230230

To submit samples, researchers will provide sample DNA, with or without primers, in 1.5ml Eppendorf tubes or in capped, full skirt, PCR plates. Usually, the samples, together with a completed **Sequencing Request Form** (see attached), will be brought to the **RIOH Sequencing Room ON6012**. Samples are placed in the small freezer just inside the room and the Request Forms are left in a mailbox on the wall outside of the room **During the Covid-19 pandemic, samples with request forms will be brought to the CancerCare Manitoba (CCMB) west side delivery doors adjacent to the small parking area. You will NOT be allowed entrance into CCMB at the front doors.** Inform the person at the delivery door that these are sequencing samples for Don Dubik and 204-807-8534. Samples will be shipped out as soon as 20+ are accumulated. Once shipped, results will be emailed directly to the researcher 2 - 4 days later.

With our current sample numbers, we are usually making 2 shipments weekly; however, if sample numbers are high, more weekly shipments are made. For the past 6 months our typical turnaround has been 3-4 working days.

For more information contact me directly.

Sincerely,

Don Dubik Ph.D., Sequencing Director
675 McDermot Ave. Rm ON6008
Ph. 204-787-4146
Don.Dubik@umanitoba.ca



Submitted by: _____ Phone No.* _____
 UM Account to bill* _____ (format : xxxxxx-3xxxx-70610x-xxxx)
 Account Investigator* _____ **Investigator Signature*** _____
 Dept. _____ Bldg: _____ Rm# _____ Date* (mm/dd/yy) ___/___/___
 Email results to*: _____ *Mandatory

Sample Submission Guidelines (Tubes or Plates)

Tubes- Use 1.5mL (large) Eppendorf tubes. Label cap of each tube with sample name(name initials & a number eg. DD12)
Plates- Use 96-well, v-bottom, full skirt PCR plates capped** Minimum 24 samples. G12&H12 empty for controls.
Express Plates- Plates as above. Template & CUSTOM primer or 10ul ddH₂O (for empty wells). G12&H12 empty for controls
 Mandatory Sample names will be used. Specify template type and size.
PowerRead Tubes (for difficult samples)- Keep sample 12ul template and 10ul primer in separate tubes.

Template Provided	Template Concentration (↑ with ↑ template size)	Custom Primer Concentration	Volume Template Only (*5 µL min. vol.)	Volume Template + Primer (*8 µL min. tube volume)
Plasmid Normal(<20kbp)	150-400 ng / µL	2 µM (pmol / µL)	10 µL (@150-300ng/µL)	10 µL + 5 µL (@2µM)
PCR (<300bp-Purified)	10-20 ng / µL	2 µM (pmol / µL)	10 µL (@10-20ng/µL)	10 µL + 5 µL (@2µM)
PCR (300-1000bp-Purified)	20-40 ng / µL	2 µM (pmol / µL)	10 µL (@20-40ng/µL)	10 µL + 5 µL (@2µM)
PCR (>1000bp-Purified)	30-60 ng / µL	2 µM (pmol / uL)	10 µL (@30-60ng/µL)	10 µL + 5 µL (@2µM)

**If you are short on template, 8 µL is the minimal tube volume needed, use ddH₂O if necessary. Tubes with volumes < 8 ul will be rejected. (NEW)*

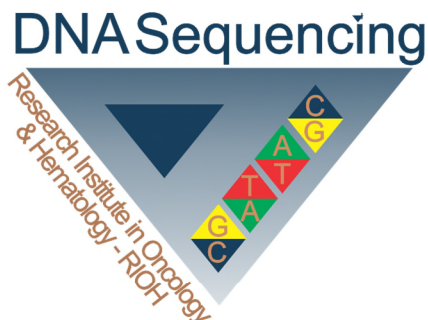
Tube #	Sample Name * (New) (*mandatory format is your name initials + no.)	Eurofin's Primer Code or CUSTOM if your primer	Template Type PCR or Plasmid	Template Size (New)	Standard or PowerRead (New)
	Eg. Sam Xu, sample 200 SX-200	Eg. T3 or T7 or SP6 or CUSTOM	Eg. PCR or Plasmid	Eg 200bp or 4.5Kb	Eg: PowerRead
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During Covid-19 bring or ship samples and completed form(s) to **CCMB, RIOH Sequencing, 675 McDermot Ave., delivery doors west side off small parking area**. Tell attendant these are sequencing samples for Don Dubik who can be reached at the number below. Shipments will be made once 20+ samples are accumulated. Cut off time for shipments is 1PM. Inclusive pricing: Tubes, \$5.50ea; Plate (24+) \$3.75ea; PowerRead Tubes, \$8.50ea and Express Plate of 94 samples, \$250. Results emailed (or placed on internal server) 24 days after shipment. All tubes must be 1.5ml Eppendorf and plates with caps must be **FisherBrand Cat# 14230237 & 14230230. For fastest delivery from Fort Garry Campus use UM Truck Messenger Service (see attached info). Have a question call Dr. Dubik 204-807-8534.

RIOH SEQUENCING 75+ FREE PRIMERS

Primers available in-house at no additional cost. Enter the **Primer Code** on the **Sequencing Request Form**

	Primer Code	Primer sequence 5' - 3'		Primer Code	Primer sequence 5' - 3'
1	16F	CGG TTA CCT TGT TAC GAC TT	40	petup	ATG CGT CCG GCG TAG A
2	16R	AGA GTT TGA TCC TGG CTC AG	41	pexF	GGA GCA GAC AAG CCC GTC AGG
3	3AOX	GCA AAT GGC ATT CTG ACA TCC	42	pexR	CAG GCT TTA CAC TTT ATG CTT CCG GC
4	5AOX	GAC TGG TTC CAA TTG ACA AGC	43	pFBACf	TCC GGA TTA TTC ATA CCG TCC C
5	96gIII	CCC TCA TAG TTA GCG TAA CG	44	pFBACr	CCT CTA CAA ATG TGG TAT GGC TG
6	BGHrev	TAG AAG GCA CAG TCG AGG	45	pGexF	ATA GCA TGG CCT TTG CAG G
7	CMVf	CGC AAA TGG GCG GTA GGC GTG	46	pGexR	GAG CTG CAT GTG TCA GAG G
8	CYC1Reverse	GCG TGA ATG TAA GCG TGA C	47	pGL3for	CTA GCA AAA TAG GCT GTC CC
9	EF-1aForward	TCA AGC CTC AGA CAG TGG TTC	48	pGLfor	GTA TCT TAT GGT ACT GTA ACT G
10	EGFPC1F	GAT CAC TCT CGG CAT GGA C	49	pGLrev	CTT TAT GTT TTT GGC GTC TTC C
11	EGFPC1R	CAT TTT ATG TTT CAG GTT CAG GG	50	pJET12F	CGA CTC ACT ATA GGG AGA GCG GC
12	EGFPN1F	GTC GTA ACA ACT CCG CCC	51	pJET12R	AAG AAC ATC GAT TTT CCA TGG CAG
13	EGFPN1R	GTC CAG CTC GAC CAG GAT G	52	pJET1F	GCC TGA ACA CCA TAT CCA TCC
14	GadFor	GGG ATG TTT AAT CCA CTA C	53	pJET1R	GCA GCT GAG AAT ATT GTA GGA GAT C
15	GadRev	AAG AAA TTG AGA TGG TGC AC	54	pJETSEQF	CGA CTC ACT ATA GGG AGA GCG GC
16	Gal4AD	TAC CAC TAC AAT GGA TG	55	pJETSEQR	AAG AAC ATC GAT TTT CCA TGG CAG
17	Gal4BD	TCA TCG GAA GAG AGT AG	56	pQEfor	GTA TCA CGA GGC CCT TTC GTC T
18	GAPSEQF	GTT CGA CCC CGC CTC GAT CC	57	pQErev	CAT TAC TGG ATC TAT CAA CAG GAG
19	GAPSEQR	TGA CAC ACA TTC CAC AGG GTC	58	pShCMVf	GGT CTA TAT AAG CAG AGC TG
20	GL2	CTT TAT GTT TTT GGC GTC TTC CA	59	pShCMVr	GTG GTA TGG CTG ATT ATG ATC AG
21	ITS1	TCC GTA GGT GAA CCT GCG G	60	RV3	CTA GCA AAA TAG GCT GTC CCC
22	ITS4	TCC TCC GCT TAT TGA TAT GC	61	SeqL-A	TCG CGT TAA CGC TAG CAT GGA TCT C
23	M1396	TGA GTT TCG TCA CCA GTA	62	SeqL-B	GTA ACA TCA GAG ATT TTG AGA CAC
24	M13F	TGT AAA ACG ACG GCC AGT	63	SP6	CAT TTA GGT GAC ACT ATA G
25	M13F43	AGG GTT TTC CCA GTC ACG ACG TT	64	SV40pA-R	GAA ATT TGT GAT GCT ATT GC
26	M13R	CAG GAA ACA GCT ATG ACC	65	SV40-promoter	TAT TTA TGC AGA GGC CGA GG
27	M13R49	GAG CGG ATA ACA ATT TCA CAC AGG	66	T3	AAT TAA CCC TCA CTA AAG GG
28	malE	GGT CGT CAG ACT GTC GAT GAA GCC	67	T7	TAA TAC GAC TCA CTA TAG GG
29	mCherry-F	CCC CGT AAT GCA GAA GAA GA	68	T7EEV	ATG TCG TAA TAA CCC CGC CCC G
30	mCherry-R	TTG GTC ACC TTC AGC TTG G	69	T7PCS2	TGT CTG GAT CTA CGT AAT ACG
31	Myc	GCA TCA ATG CAG AAG CTG ATC TCA	70	T7term	CTA GTT ATT GCT CAG CGG T
32	pBabe3	ACC CTA ACT GAC ACA CAT TCC	71	TrcHisF	AAT CTG TGT GGG CAC TCG
33	pBabe5	CTT TAT CCA GCC CTC AC	72	TrcHisR	CTT CTG CGT TCT GAT TTA ATC TG
34	pBabeF	TGA CCT GGG AAG CCT TGG CT	73	V1-5	GGA CTT TCC AAA ATG TCG
35	pBabeR	TTG CTG ACT AAT TGA GAT GCA TGC TTT	74	V5	ACC GAG GAG AGG GTT AGG GA
36	pBadF	ATG CCA TAG CAT TTT TAT CC	75	VF2	TGC CAC CTG ACG TCT AAG AA
37	pBadR	GAT TTA ATC TGT ATC AGG	76	VR	ATT ACC GCC TTT GAG TGA GC
38	pcDNAF	GGC TAA CTA GAG AAC CCA CTG	77	XL39	ATT AGG ACA AGG CTG GTG GG
39	pcDNAR	GGC AAC TAG AAG GCA CAG TC		CUSTOM	Your provided primer premixed with the template



Free delivery from Fort Garry to Bannatyne Campus

The only current option for free delivery of samples between the campuses is Inter-departmental Mail.

Unfortunately this service take about a week to deliver.

Paid (Not Free) Options:

1)University of Manitoba Truck Messenger Service \$20 flat rate

The UofM has a truck messenger service that delivers items between the Fort Garry and Bannatyne campuses. This service is provided by Physical Plant and includes pickup from your departmental office and delivery to the other campus, usually within the same day. They have recently started to enforce their Policies, Section 2.4 that states:

“Organizations or individuals which are funded for research purposes, and those organizations which are income producing, are responsible for delivery services used to support research or income producing activities”.

http://umanitoba.ca/admin/financial_services/secure/truck_messenger.php

Need more info, contact 204 - 474 - 8911.

Procedure for sending sample to RIOH DNA Sequencing

Prepare your samples and complete the RIOH Sequencing Request Form available at

<https://www.dropbox.com/sh/tx99tf0atp4w4fz/AACSPfUNbSiU58th8je0Peea?dl=0>

Place the sample tubes and the MICB Sequencing Request Form into a sturdy envelope or box. Sample should be send as is, without ice.

Address the envelope to:

**RIOH Sequencing
c/o Dr. Don Dubik
675 McDermot Ave Rm ON6008
Wpg, MB R3E0V9
ph. 204 -787 - 4146**

Samples can be delivered or brought to the CancerCare Manitoba delivery doors 675 McDermot Ave. on the west side adjacent to the small parking area. Walk through the delivery doors and leave your samples on the table next to the door.

Inform the person at the delivery door that these are sequencing samples for Don Dubik and 204 807 - 8534.

Samples will be shipped out as soon as 20+ are accumulated. Once shipped, results will be emailed directly to the researcher 2 - 4 days later