

Ebola virus sequencing kit-list

Nanopore | amplicon | native barcoding

Document: ARTIC-EBOV-seqKit-v1.0.0

Creation Date: 2018-05-21

Revision Date: 2018-05-21

Forked from: doi:10.1038/nprot.2017.066

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Overview: The following protocol is adapted from the methods of Quick et al. (2017) *Nature Protocols* **12:** 1261–1276 doi:10.1038/nprot.2017.066 **2** and covers primers, amplicon preparation and clean-up, then uses a single-tube protocol to barcode and adaptor ligate the library, before running minION.

Ebola primer scheme:

https://github.com/artic-network/primer-schemes/tree/master/ZaireEbola/V2 2

Ebola virus Nanopore sequencing protocol:

http://artic.network/ebov/ebov-seq-sop-1.0.0.html

Ebola virus Nanopore sequencing kit-list:

http://artic.network/ebov/ebov-seq-kit-1.0.0.html



Funded by the Wellcome Trust

Collaborators Award 206298/Z/17/Z --- ARTIC network (artic.network)

Library Preparation and Sequencing Reagents:

Product	Supplier	Cat No.	Unit Cost	# Req	Cost	
Vilo SSIV RT	Thermo	11756050	£793	1	£793	
Q5® Hot Start HF Polymerase (2xMM)	NEB	M0494L	£611	1	£611	
NEBNext® Ultrall™ End-prep	NEB	E7456L	£722	1	£722	
Ebola Zaire V2 Primers*	Thermo		£240	1	£220	
NEBNext® Ultrall™ Ligation Module	NEB	E7595L	£1,204	1	£1,204	
NEBNext® FFPE Repair Module	NEB	M6630L	£539	1	£539	
Agencourt® AmpureXP™ Beads (60mL)	FisherSci	10453438	£620	1	£620	
Ligation Sequencing Kit 1D	Nanopore	SQK-LSK108	£450	4	£1,800	
Low Input by PCR Barcoding Kit	Nanopore	SQK-LWB001	£500	2	£1,000	
Native Barcoding Expansion Kit	Nanopore	EXP-NBD103	£250	4	£1,000	
R9.4.1 flow cells	Nanopore		£375	12	£4,500	
Qubit® dsDNA HS Assay Kit	Thermo	Q32851	£185	2	£370	
TOTAL COST					£13,379	

Equipment and Containment

Product	Supplier	Cat No.	Unit Cost	# Req	Cost	
1000µL pipettes	Gilson	10387322	£210	2	£420	
100μL pipettes	Gilson	10442412	£210	2	£420	
10µL pipettes	Gilson	10236072	£210	2	£420	
Sprout Microfuge (12V)	PhilipHarris	B8R04574	£170	2	£340	
PV-1 Mini Vortex	WolfLabs	PV-1	£150	2	£300	
Portable Heat Block	Mobitec	Pad3-H-CB	£310	1	£310	
DynaMag-2 magnetic rack	Thermo	12321D	£540	1	£540	
MiniPCR™ Mini8	Cambio	MPQP100001	£535	4	£2,140	
MyGo® Mini qPCR	IT-IS	SKU7642	£6,500	1	£6,500	
Qubit4® spectrophotometer	Thermo	Q33228	£2,578	1	£2,578	
Collapsible Tube racks (small x5)	FisherSci	11770045	£50	1	£50	
Collapsible Tube racks (large x5)	FisherSci	11710634	£50	1	£50	
Cold Racks (0.2-0.5mL)	Sigma	Z606634-1EA	£50	2	£100	
Lab Mats	StarLab	E3860-0061	£25	2	£50	
USB Lights	Amazon	Jebsen-T05	£20	2	£40	
Portable UV Lamps	Amazon	B078LSR3MG	£20	2	£40	
Portable glove box	Amazon	B01B1MGRR0	£70	2	£40	
TOTAL COST					£14,438	

Consumables and Disposables

Product (Number)	Supplier	Cat No.	Unit Cost	# Req	Cost	
Stacked 1000µL Filter Tips (500)	FisherSci	11977724	£70	2	£140	
Stacked 100µL Filter Tips (1000)	FisherSci	11947724	£60	1	£60	
Stacked 10µL Filter Tips (1000)	FisherSci	11907724	£60	1	£60	
1.5mL Eppendorf Tubes (500)	StarLab	I1415-5500	£20	2	£40	
0.6mL Eppendorf Tubes (500)	StarLab	I1405-1500	£20	1	£20	
0.2mL 8-strip tubes (1000)	StarLab	I1402-3600	£60	1	£60	
Qubit Reaction Tubes (500)	Thermo	Q32856	£50	1	£50	
MyGo PCR Tubes (500)	IT-IS	SKU7688	£85	1	£85	
BioBln® Disposal Bins (100)	FisherSci	10394373	£121	1	£121	
Gloves (Large, 100)	StarLab	SG-P-L	£5	2	£10	
Gloves (Small, 100)	StarLab	SG-P-S	£5	2	£10	

Product (Number)	Supplier	Cat No.	Unit Cost	# Req	Cost	
Paper Towels	Generic			4	N/A	
Decon Wipes (240)	MediPal	PMC6038	£6	2	£12	
Back-tie Lab Gowns (45G)	VWR	113-8316	£56	10	£56	
RNAse Away (250mL)	Thermo	10328011	£60	2	£120	
DNA Away (250mL)	Thermo	7010PK	£20	2	£40	
TOTAL COST					£884	

Cold Chain, Power and Storage

Product (Number)	Supplier	Cat No.	Unit Cost	# Req	Cost	
Flat dehydrated cold sheets (ThermaFreeze)	Amazon	B003ET2G74	£15	10	£150	
Thermos Radiance Cooler Bag	Amazon	B00BHW5B4U	£10	4	£40	
Portable Fridge Bag	Amazon	B008N5UU3G	£70	1	£70	
Credo Cube Series 20M 4L	Pelican	Contact Supp.			N/A	
PowerAdd Pilot Pro2 85Wh LilON 9-20V	Amazon	B013HXKZYW	£80	2	£160	
Solar Charger, 85Wh, LilON 5-20V	Amazon	B00AE9FBCO	£85	2	£170	
Goal Zero Yeti 150 Portable Power Station	GoalZero	SKU22004	£200	1	£200	
Goal Zero Nomad 28 Plus Solar Panel	Goal Zero	SKU11805	£250	1	£250	
Surge Protector Extension (4-plug)	Belkin	B0002AG0F2	£6	3	£18	
SKROSS Universal Mini Travel Adaptor	Amazon	B00OHSWG6C	£10	2	£20	
Peli Air 1615 Waterproof Equipment Case	R&S Comp.	121-8893	£550	1	£550	
Eagle Creek Cargo Hauler, 120L	Amazon	B01HIGO3OC	£150	1	£150	
BESTEK 200W Power Inverter DC 12V	Amazon	B01N2P9V64	£30	1	£30	
Samsung Portable SSD T5 1TB	Amazon	B074M774TW	£320	2	£640	
Seagate® 5TB External HD	Amazon	B01M058Q39	£119	2	£238	

TOTAL COST £2,686