

Consumables and Equipment

Check to make sure that you have all of the necessary user-supplied consumables and equipment before proceeding to the TruSeq Synthetic Long-Read DNA Library Prep protocol.



NOTE

The TruSeq Synthetic Long-Read DNA Library Prep protocol has been optimized and validated using the items listed. Comparable performance is not guaranteed when using alternate consumables and equipment.

Table 4 User-Supplied Consumables

Consumable	Supplier
10 μ l barrier pipette tips	General lab supplier
10 μ l multichannel pipettes	General lab supplier
10 μ l single channel pipettes	General lab supplier
1000 μ l barrier pipette tips	General lab supplier
1000 μ l multichannel pipettes	General lab supplier
1000 μ l single channel pipettes	General lab supplier
20 μ l barrier pipette tips	General lab supplier
20 μ l multichannel pipettes	General lab supplier
20 μ l single channel pipettes	General lab supplier
200 μ l barrier pipette tips	General lab supplier
200 μ l electronic eight-channel pipette	General lab supplier
200 μ l multichannel pipettes	General lab supplier
200 μ l single channel pipettes	General lab supplier
1 Kb DNA Extension Ladder	Invitrogen, part # 10511-012

Consumable	Supplier
2-Log DNA Ladder (0.1–10.0 kb)	NEB, part # N3200L
2-propanol (Isopropanol)	General lab supplier
15 ml conical tube	General lab supplier
15 ml RNase/DNase-free reagent reservoirs	General lab supplier
50 ml conical tube	General lab supplier
96-well 0.3 ml skirtless PCR plates or Twin.Tec 96-well PCR plates	E&K Scientific, part # 480096 Eppendorf, part # 951020303
96-well storage plates, round well, 0.8 ml ("MIDI" plate)	Fisher Scientific, part # AB-0859
Axygen 384 well PCR microplate	VWR, part # 10011-194 or 47744-810
Compression Mats for PCR Plates	VWR, part # 10011-006
Dimethyl sulfoxide (DMSO)	General lab supplier
DNA Binding Buffer	Zymo Research, part # D4003-1-25
DNA Wash Buffer	Zymo Research, part # D4003-2-24
E-Gel EX Agarose Gels, 1%	Invitrogen, catalog # G4020-01
E-Gel NGS 0.8% Agarose Gels	Invitrogen, catalog # A25798
Ethanol 200 proof (absolute) for molecular biology (500 ml)	Sigma-Aldrich, part # E7023
g-TUBE	Covaris, part # 520079
Ice bucket	General lab supplier
KAPA Library Quantification Kit - Illumina/Universal	KAPA Biosystems, part # KK4824

Consumable	Supplier
Lab pen	General lab supplier
Lab tissue, low-lint	VWR, part # 21905-026 or equivalent
Microcentrifuge Tubes with Attached Flat Caps, Neptune (1.6 ml)	VWR, part # 89126-722
Microseal 'B' adhesive seals	Bio-Rad, part # MSB-1001
Needles (22 1/2 gauge)	General lab supplier
PCR-grade water	General lab supplier
QIAquick Gel Extraction Kit	QIAGEN, part # 28704
qPCR plate and seal	General lab supplier
Qubit dsDNA BR Assay Kit	Life Technologies catalog # Q32850
Qubit dsDNA HS Assay Kit	Life Technologies catalog # Q32851
RNase/DNase-free eight-tube strips and caps	General lab supplier
RNase/DNase-free multichannel reagent reservoirs, disposable	VWR, part # 89094-658
RNaseZap (to decontaminate surfaces)	General lab supplier
ROX Reference Dye	Invitrogen, part # 12223012
Ruler	General lab supplier
SYBR Green Nucleic Acid Gel Stain	Invitrogen, part # S7585
x-tracta gel extractor	USA Scientific, catalog # 5454-0100

Consumable	Supplier
Zymo-Spin V with Reservoir	Zymo Research, part # C1016-25 or C1016-50
[Optional - for BluePippin size selection] 0.75% Agarose gel cassettes , Dye Free, Low Range 10/pk S1	Sage Science, catalog # BLF7510
[Optional - for BluePippin size selection] TE buffer	General lab supplier

Table 5 User-Supplied Equipment

Equipment	Supplier
96-well thermal cycler with heated lid	Bio-Rad, part # ALS-1296G or equivalent
384-well thermal cycler	Bio-Rad, part # 185-1138 or equivalent
2100 Bioanalyzer Desktop System	Agilent, part # G2940CA
Agilent High Sensitivity DNA Kit	Agilent, part # 5067-4626
Dark reader transilluminator	Clare Chemical Research, part # DR195M
E-Gel iBase Power System	Life Technologies, catalog # G6400
High-Speed Microplate Shaker	VWR, catalog # 13500-890 (110 V/120 V) VWR, catalog # 14216-214 (230 V)
Magnetic stand-96	Life Technologies, catalog # AM10027
Microplate centrifuge	General lab supplier
Novex Gel Knife	Life Technologies catalog # EI9010

Equipment	Supplier
qPCR system See <i>qPCR Systems</i> on page 87.	General lab supplier
Qubit 2.0 Fluorometer	Life Technologies, catalog # Q32866
Vacuum manifold	Promega, catalog # A7231 or QIAGEN, part # 19413
[Optional - for BluePippin size selection] BluePippin Size Selection System	Sage Science, catalog # BLU0001
[Optional - for SYBR Green calibration] NanoDrop Spectrophotometer	Thermo Scientific, catalog # ND-1000 or ND-2000

qPCR Systems

The following table lists the validated qPCR systems for the TruSeq Synthetic Long-Read DNA Library Prep protocol. Either a 96-well or 384-well qPCR system is required.

Equipment	Supplier
CFX96 Touch Real-Time PCR Detection System ¹	Bio-Rad, part # 185-5195
CFX384 Touch Real-Time PCR Detection System	Bio-Rad, part # 185-5484
7900HT Fast Real-Time PCR System with 384-Well Block Module ²	Life Technologies, part # 4329001
Mx3000P qPCR System	Agilent, part # 401511
LightCycler 480 Instrument II (384-well version)	Roche, part # 05015243001

1. Illumina recommends using CFX Manager software version 3.0 with Cq Determination mode: Single Threshold; Baseline Setting: Baseline Subtracted Curve Fit and Apply Fluorescent Drift Correction for data analysis. This can correct for abnormalities in fluorescence intensity of the standard curve caused by the instrument. For software installation, contact Bio-Rad.
2. When setting up the SDS software before running the qPCR, select only the wells with samples. If the entire plate is selected, the resulting file is too large to open by the SDS software. For more information, contact Life Technologies.