

Materials List for:**Discrimintion and Mapping of the Primary and Processed Transcripts in Maize Mitochondrion Using a Circular RT-PCR-Based Strategy**Yafeng Zhang^{1,2}, Xun Liao², Peihua Luo², Kui Peng², Wenyi Ye², Tengxiang Lian^{1,2}, Qibin Ma^{1,2}, Hai Nian^{1,2}¹State Key Laboratory of Conservation and Utilization of Subtropical Agro-Bioresources, South China Agricultural University²Guangdong Provincial Key Laboratory of Plant Molecular Breeding, College of Agriculture, South China Agricultural UniversityCorrespondence to: Yafeng Zhang at yafeng_zhang@scau.edu.cn, Hai Nian at hnian@scau.edu.cnURL: <https://www.jove.com/video/60019>

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Materials

Name	Company	Catalog Number	Comments
Acetic acid	Aladdin, China	A112880	To prepare 1x TAE buffer
Applied Biosystems 2720 Thermal Cycler	Thermo Fisher Scientific, USA	4359659	Thermal cycler for PCR amplification
Ascorbic acid	Sigma-aldrich, USA	V900134	For preparation of extraction buffer
Biowest Agarose	Biowest, Spain	9012-36-6	To resolve PCR products and RNAs
Bovine serum albumin	Sigma-aldrich, USA	A1933	For preparation of extraction buffer
Bromophenol blue	Sigma-aldrich, USA	B8026	For preparation of loading buffer for agarose gel electrophoresis and Northern blot
DEPC	Sigma-aldrich, USA	V900882	Deactivation of RNase
DIG Northern starter kit	Roche, USA	12039672910	For DIG-RNA labeling and Northern blot. This kit contains the reagents for transcription-labeling of RNA with DIG and T7 RNA polymerase, hybridization and chemiluminescent detection.
EDTA	Sigma-aldrich, USA	V900106	For preparation of extraction buffer and 1x TAE buffer
EGTA	Sigma-aldrich, USA	E3889	For preparation of wash buffer
Gel documentation system	Bio-Rad, USA	Gel Doc XR+	To image the agarose gel
Glycerol	Sigma-aldrich, USA	G5516	For preparation of loading buffer for agarose gel electrophoresis
GoldView II (5000x)	Solarbio, China	G8142	DNA staining
Hybond-N+, Nylon membrane	Amersham Biosciences, USA	RPN119	For Northern blot
Image Lab	Bio-Rad, USA	Image Lab 3.0	Image gel, and compare the abundance of PCR products.
KH ₂ PO ₄	Sigma-aldrich, USA	V900041	For preparation of extraction buffer
KOH	Aladdin, China	P112284	For preparation of extraction buffer
L-cysteine	Sigma-aldrich, USA	V900399	For preparation of extraction buffer
Millex	Millipore, USA	SLHP033RB	To sterile extraction and wash buffers by filtration
Miraclot	Calbiochem, USA	475855-1R	To filter the ground kernel tissues
MOPS	Sigma-aldrich, USA	V900306	For preparation of running buffer for Northern blot
NanoDrop	Thermo Fisher Scientific, USA	2000C	For RNA concentration and purity assay
NaOH	Sigma-aldrich, USA	V900797	For preparation of wash buffer

pEASY-Blunt simple cloning vector	TransGen Biotech, China	CB111	Cloning of the gel-recovered band. It contains a T7 promoter several bps upstream of the insertion site.
Phanta max super-fidelity DNA polymerase	Vazyme, China	P505	DNA polymerase for PCR amplification
Polyvinylpyrrolidone 40	Sigma-aldrich, USA	V900008	For preparation of extraction buffer
Primer Premier 6.24	PREMIER Biosoft, USA	Primer Premier 6.24	To design primers for reverse transcription and PCR amplification
PrimeScript II reverse transcriptase	Takara, Japan	2690	To synthesize the first strand cDNA
PureLink RNA Mini kit	Thermo Fisher Scientific, USA	12183025	For RNA purificaion
RNA 5' polyphosphatase	Epicentre, USA	RP8092H	To convert 5' triphosphate to monophosphate
RNase inhibitor	New England Biolabs, UK	M0314	A component of RNA self-ligation and 5' polyphosphatase treatment reactions, and it is used to inhibite the activity of RNase.
Sodium acetate	Sigma-aldrich, USA	V900212	For preparation of running buffer for Northern blot
Sodium chloride	Sigma-aldrich, USA	V900058	To prepare 20x SSC
SsoFas evaGreen supermixes	Bio-Rad, USA	1725202	For RT-qPCR
T4 RNA Ligase 1	New England Biolabs, UK	M0437	For RNA circularization
Tetrasodium pyrophosphate	Sigma-aldrich, USA	221368	For preparation of extraction buffer
TIANGel midi purification kit	Tiangen Biotech, China	DP209	To purify DNA fragments from agarose gel
Tris	Aladdin, China	T110601	To prepare 1x TAE buffer
TRIzol reagent	Invitrogen, USA	15596026	To extract mitochondrial RNA.
Universal DNA purification kit	Tiangen Biotech, China	DP214	To recover linearized plasmids from the restriction enzyme digestion reaction
Xylene cyanol FF	Sigma-aldrich, USA	X4126	For preparation of loading buffer for agarose gel electrophoresis