

RNA isolation

Special considerations for isolation of RNA from different sample sources

Plants



Challenges

- co-purified metabolites inhibiting enzymatic reactions
- increased viscosity leading to pipetting errors
- RNA degradation during storage

Considerations

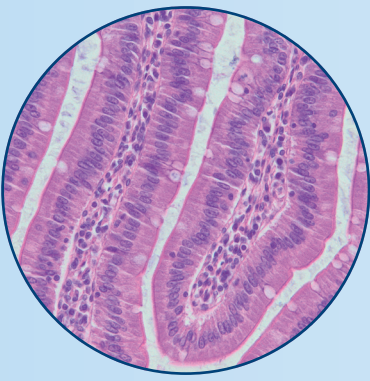
- use plants grown under conditions that do not induce high levels of metabolites (grow plants in darkness for 1 to 2 days before harvesting)
- use healthy, young tissues when possible

Recommended Kit

RNeasy Plant Mini Kit

With QIAshredder columns for easy homogenization

Heart, muscle and skin tissue



Challenges

- abundant contractile proteins, connective tissue, and collagen interfering with RNA isolation

Considerations

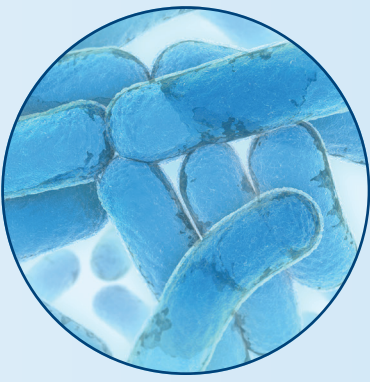
- treat sample with a protease or phenol-containing lysis reagent under conditions preventing RNA degradation

Recommended Kit

RNeasy Fibrous Tissue Mini Kit

With Proteinase K to remove structural proteins

Bacteria



Challenges

- highly unstable, rapid mRNA turnover and degradation

Considerations

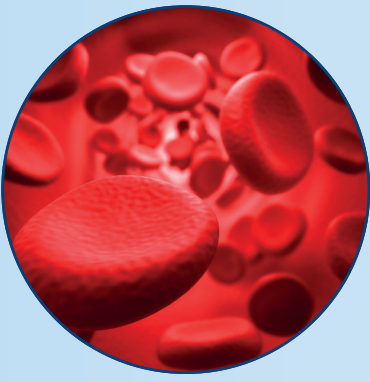
- stabilize samples prior to sample harvesting and processing

Recommended Kit

RNeasy Protect Bacteria Mini Kit

With RNAprotect Bacteria Reagent for stabilizing RNA

Blood



Challenges

- enzyme inhibitors and anticoagulants inhibiting downstream RNA analysis

Considerations

- preserve RNA using an RNA stabilization reagent in the collection tube
- remove anticoagulants and enzyme inhibitors
- remove unwanted erythrocytes by selective lysis
- use Ficoll density-gradient centrifugation to recover mononuclear cells

Recommended Kit

QIAamp RNA Blood Mini Kit

With differential lysis to remove erythrocytes; QIAshredder for homogenization

FFPE tissue samples



Challenges

- heavily fragmented and chemically modified (by formaldehyde) nucleic acids due to fixation and embedding conditions

Considerations

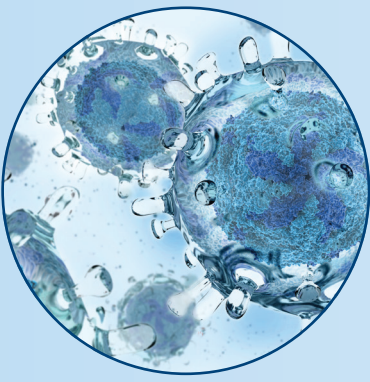
- store FFPE samples at 4°C
- remove and fix tissue quickly
- use tissue samples as thin as 5 mm and do not over-fix (max. 24 h)
- use high-quality reagents for paraffin embedding
- avoid sample staining
- use an appropriate deparaffinization step
- involve a crosslink-reversal step during RNA isolation

Recommended Kits

RNeasy FFPE Kit AllPrep DNA/RNA FFPE Kit

With efficient cross-link removal

Virus



Challenges

- low titer and a high degree of secondary structure leading to low yield and difficult downstream analysis
- high mutation rate due to inaccurate copying while replicating
- difficult to obtain a homogeneous population for analysis

Considerations

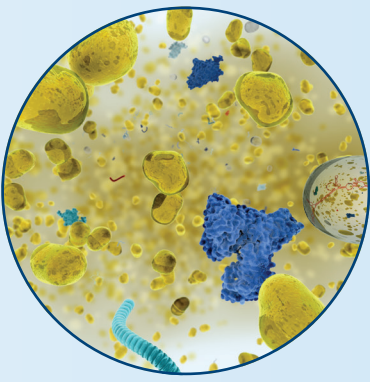
- concentrate virus particles by ultracentrifugation, ultrafiltration, or precipitation before RNA isolation
- add carrier RNA during RNA isolation

Recommended Kit

QIAamp Viral RNA Mini Kit

With optional carrier RNA for improved binding of low titer viral RNA

Other body fluids (plasma, serum, urine)



Challenges

- lower concentration and rapid degradation of RNA by repeated freeze-thaw cycles
- high amount of RNases and inhibitors interfering with downstream assays

Considerations

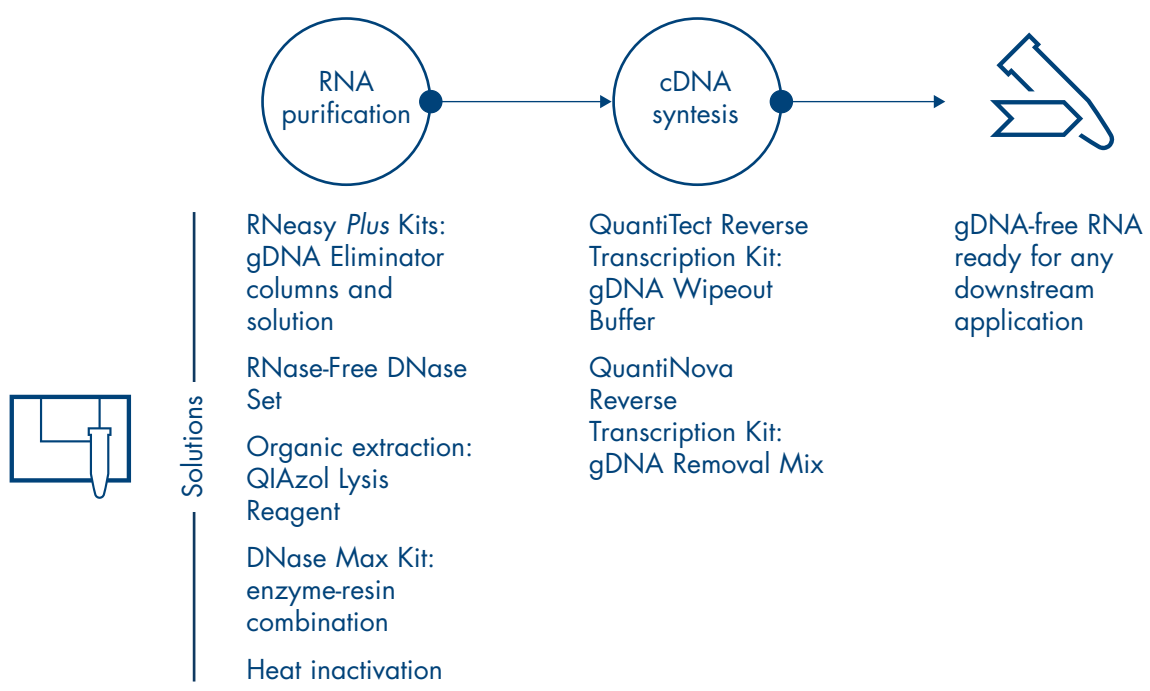
- use dedicated isolation protocols to maximize yield while avoiding degradation or inhibition

Recommended Kits

miRNeasy Serum/Plasma Advanced Kit exoRNeasy Serum/Plasma Maxi Kit

With phenol-free isolation of miRNAs from serum/plasma; easy isolation of exosomal RNA and miRNA without ultracentrifugation

Removal of genomic DNA contamination from RNA



Find a selection of RNA isolation technologies for any sample type at www.qiagen.com/SelectRNA.

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