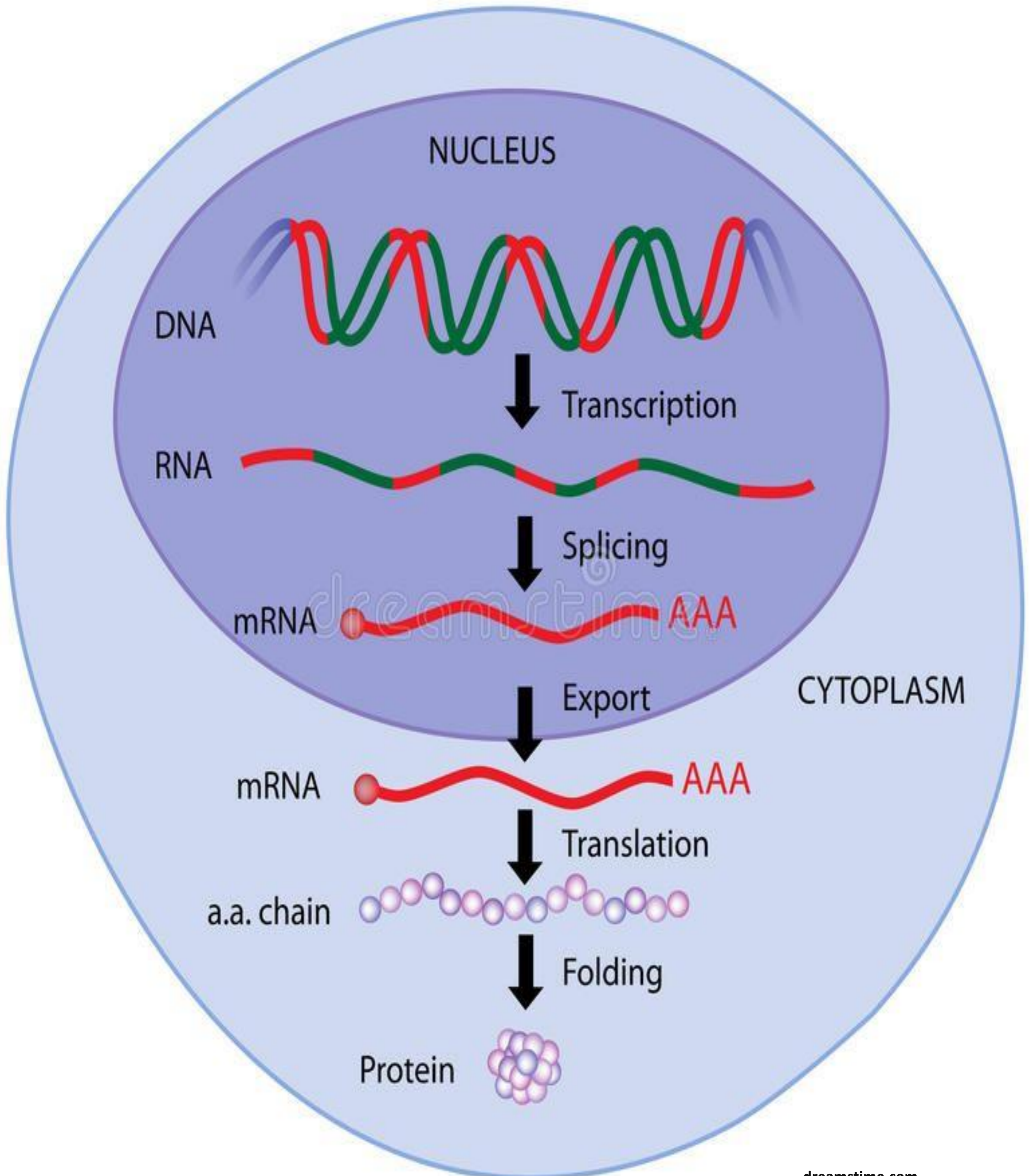


A low-angle photograph of a large, mature tree with a thick, textured trunk and a dense canopy of green leaves. The tree's branches spread out in all directions, filling most of the frame. The background is a bright, slightly overcast sky, which makes the green of the leaves stand out. The overall scene is peaceful and natural.

RNA interference and its application in the treatment of tree diseases

Central dogma of molecular biology



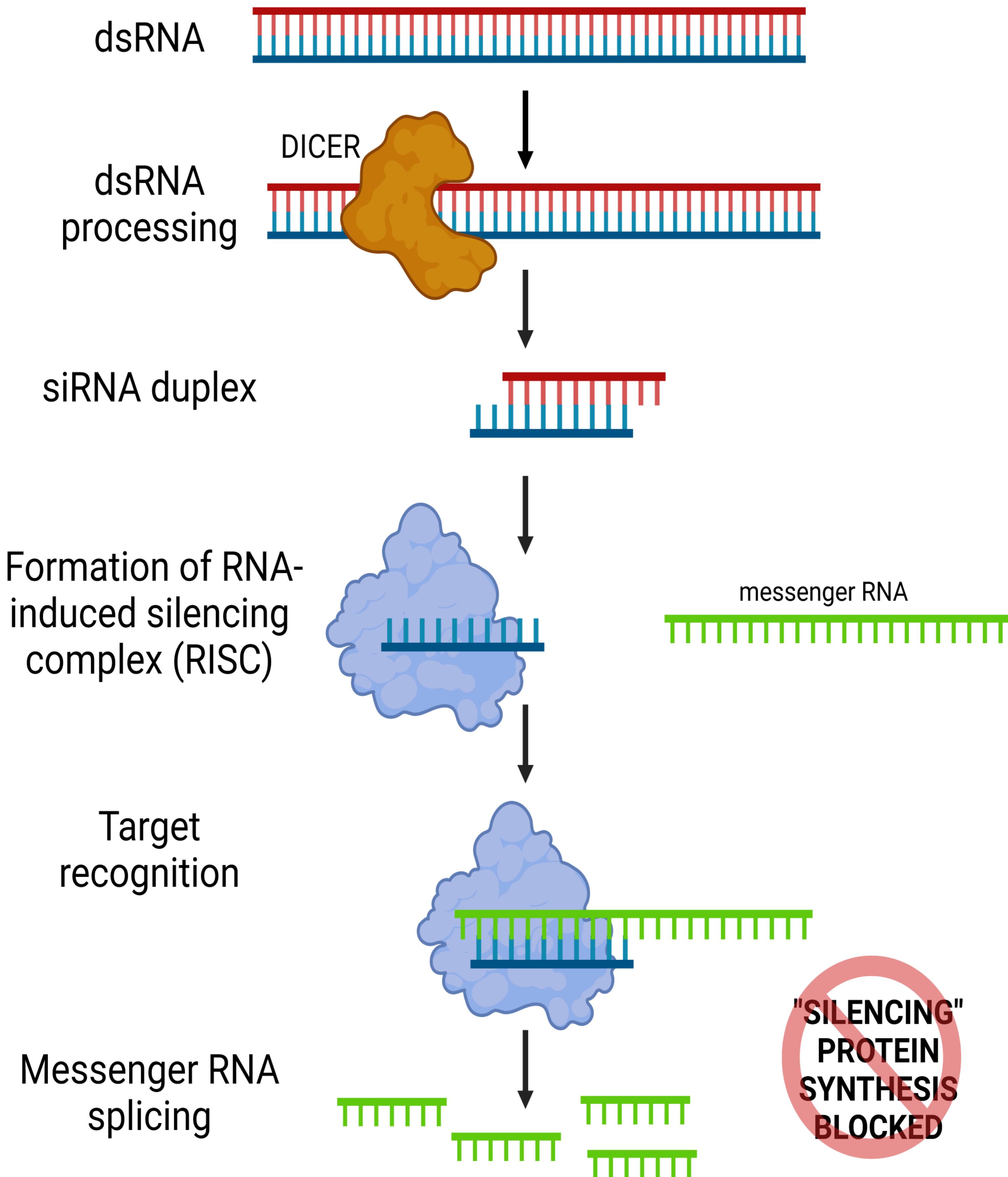
What is RNA interference (RNAi)?

- Conserved biological response to double-stranded RNA
- Mediates resistance to both endogenous parasitic and exogenous pathogenic nucleic acids
- Regulates the expression of protein-coding genes
- Natural mechanism for sequence-specific gene silencing

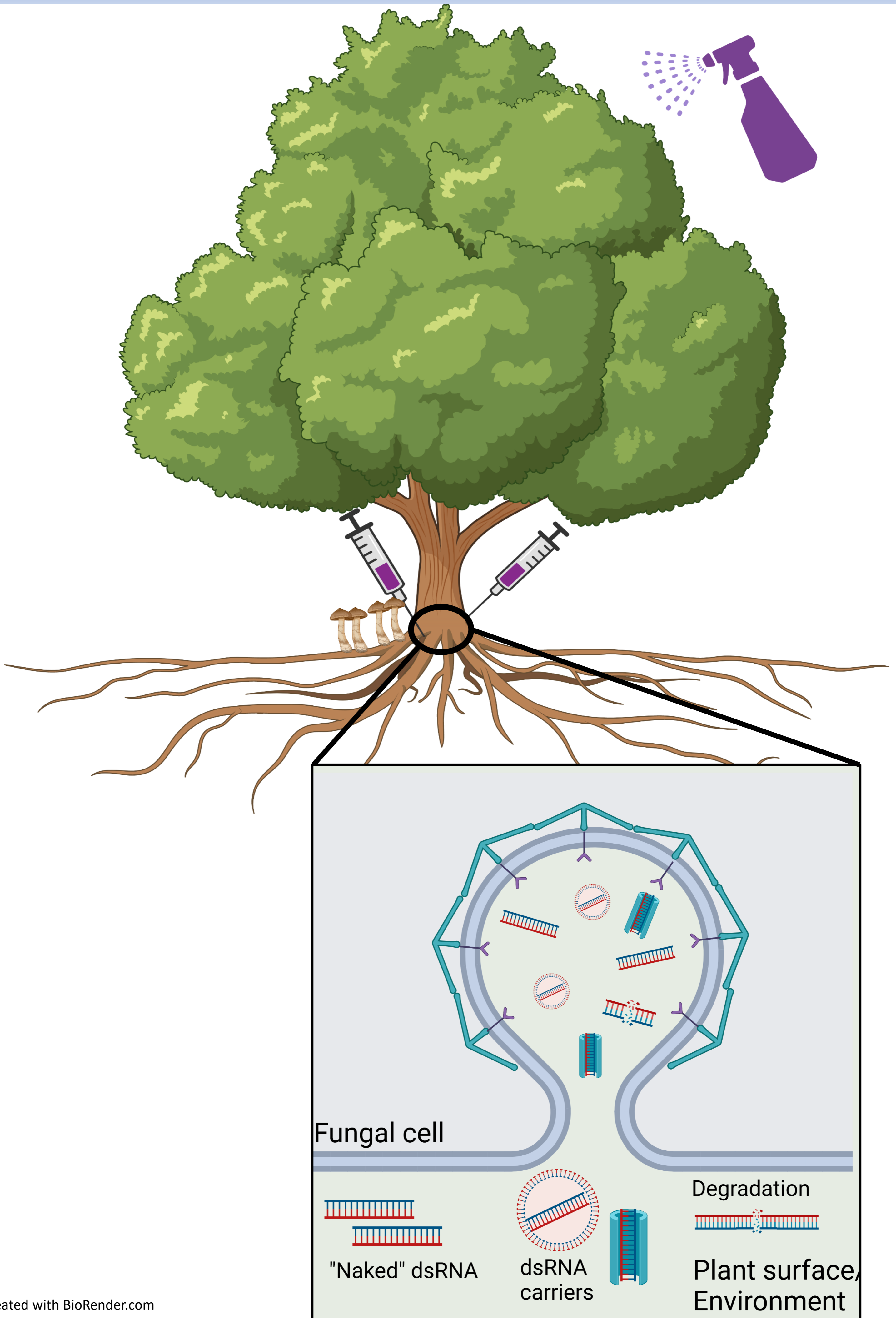
How can we exploit RNAi for biological control of pathogens?

- RNAi offers an environmentally friendly alternative to broad spectrum fungicides/pesticides to control the growth of plants pathogens and pests.
- Controls pathogens without adversely affecting non-target microorganisms or causing changes to the plant and soil microbiome
- Uses sequence specific knockdown of gene targets following delivery of double-stranded RNA
- The pathogen must internalise the dsRNA and process it via RNAi cellular machinery to silence target genes.

RNA interference



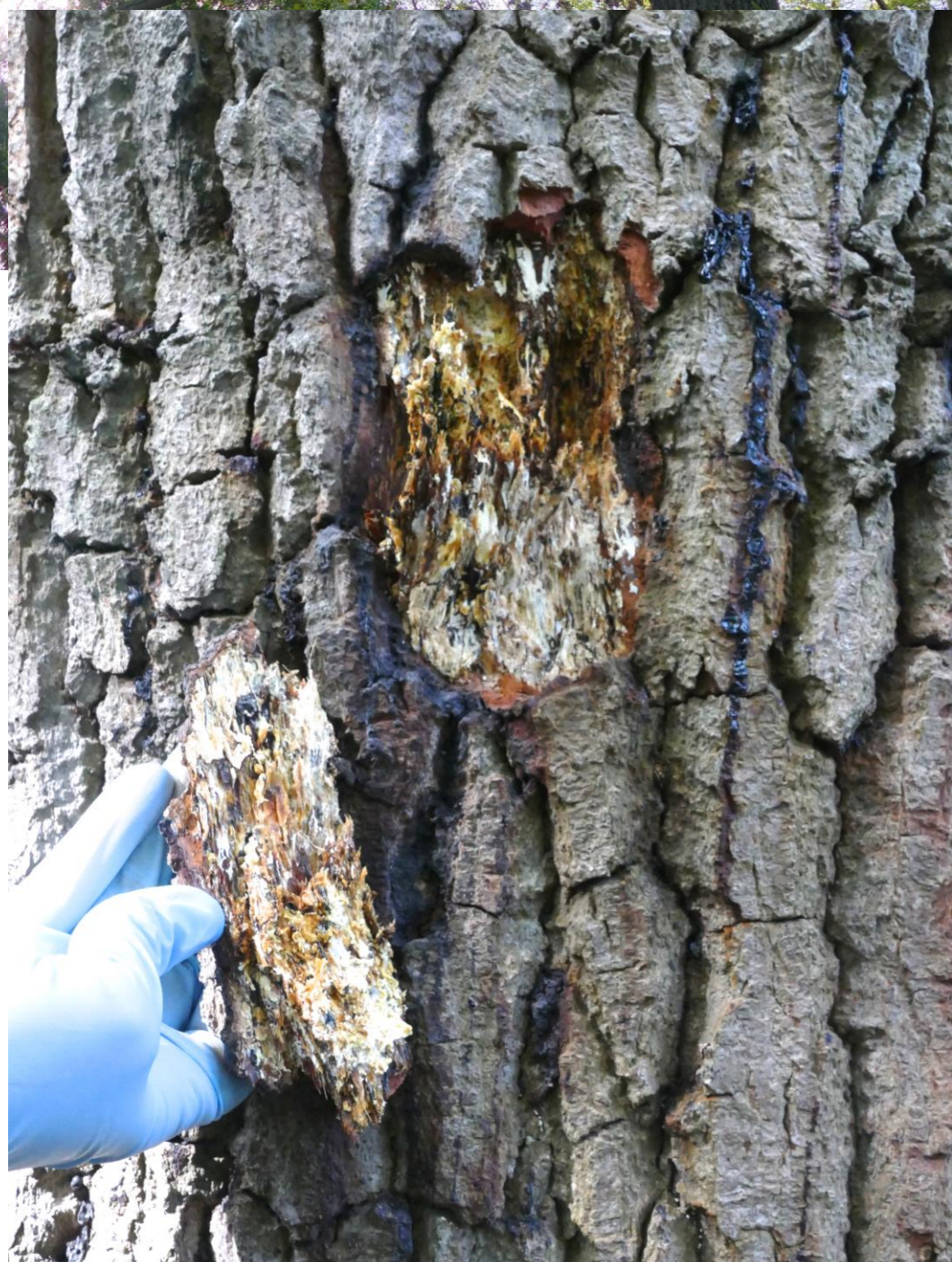
dsRNA Application and Uptake



RNAi to manage growth of *Armillaria mellea*, a major root pathogen causing decline in veteran oaks

- *Armillaria mellea*

- a major root pathogen of many parkland and forest tree species
- a predisposing factor in oak declines



How do we target *A. mellea* and not other *Armillaria* species?

Armillaria species genome comparisons

<i>A. mellea</i>	ACGGT TACCATGATCTTA TCGGATCCACTT
<i>A. gallica</i>	ACGGT TTTCAGATCCAT GGGATGGAGTCTT
<i>A. ostoyae</i>	ACGGT TTTCAGATCCAT GGGATGGAGTCTT
<i>D. tabescens</i>	ACGGT TTTCAGATCCAT GGGATGGAGTCTT

dsRNA in *A. mellea* = RNAi

dsRNA

ACGG**UACCAUGAUCUUAUC**GGAUCCACUU
UGCCAUGGU**ACUAGAAUAGCCUAGG**UGAA

siRNA

CCAUGAUCUUAUC
AUGGU**ACUAGAAU**

RISC
complex

CCAUGAUCUUAUCGGAUCCACUU
UGCCAUGGU**ACUAGAAUAGCCUAGG**UGAA

Spliced
RNA

UGCCA UGGUAC
UAGAAU AGCCUAG
GUGAA

"SILENCING"
PROTEIN
SYNTHESIS
BLOCKED

dsRNA in *A. gallica* = no RNAi

dsRNA

ACGG**UACCAUGAUCUUAUC**GGAUCCACUU
UGCCAUGGU**ACUAGAAUAGCCUAGG**UGAA

siRNA

CCAUGAUCUUAUC
AUGGU**ACUAGAAU**

RISC
complex

CCAUGAUCUUAUCGGAUCCACUU

ACGGUUUCAGAU**CCAUGGGAUGGAGUCUU**

mRNA does not bind to guide
RNA in RISC complex

Translation
& protein
synthesis

