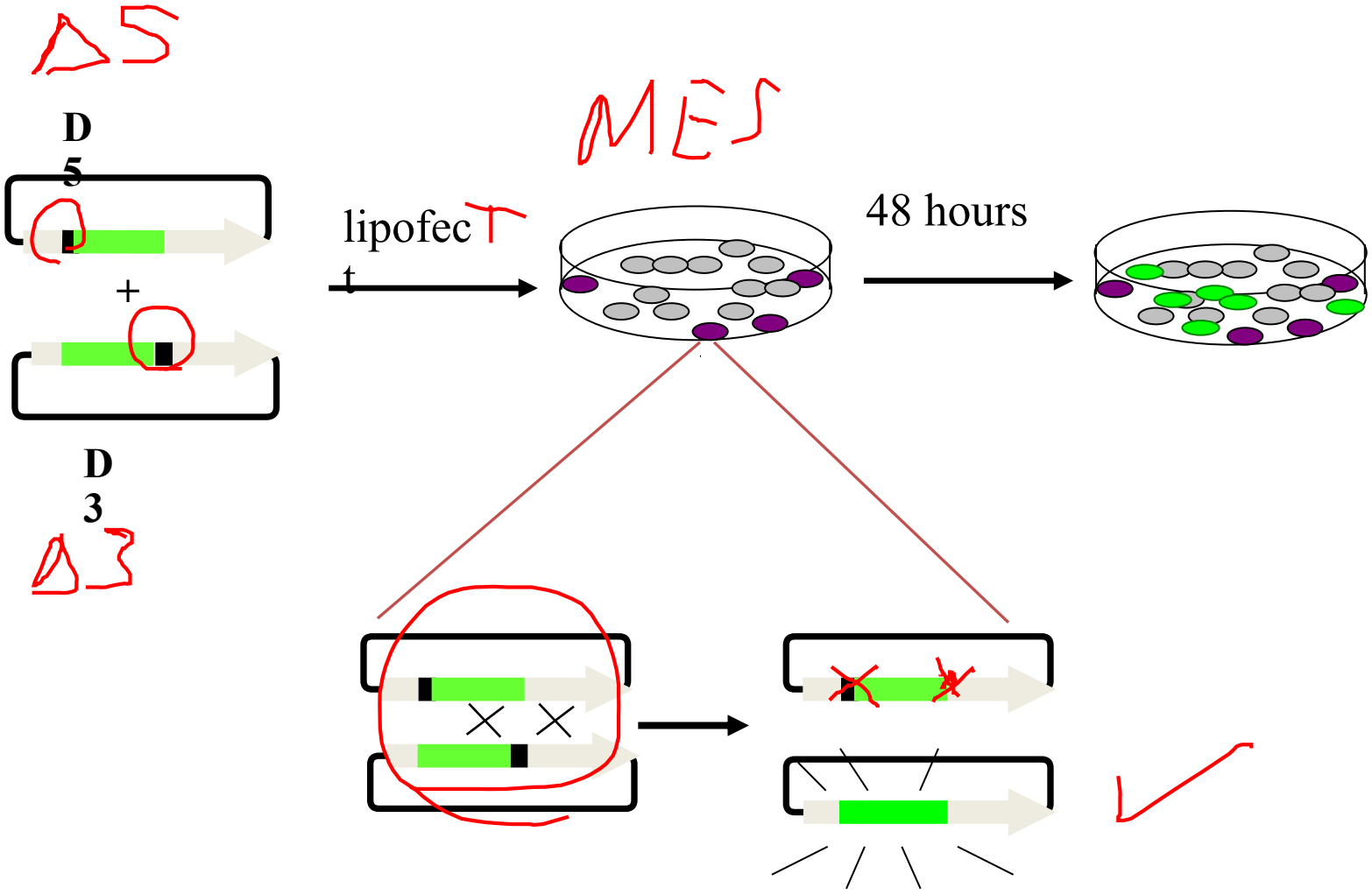
DNA Engineering: M1D1 Lab Talk

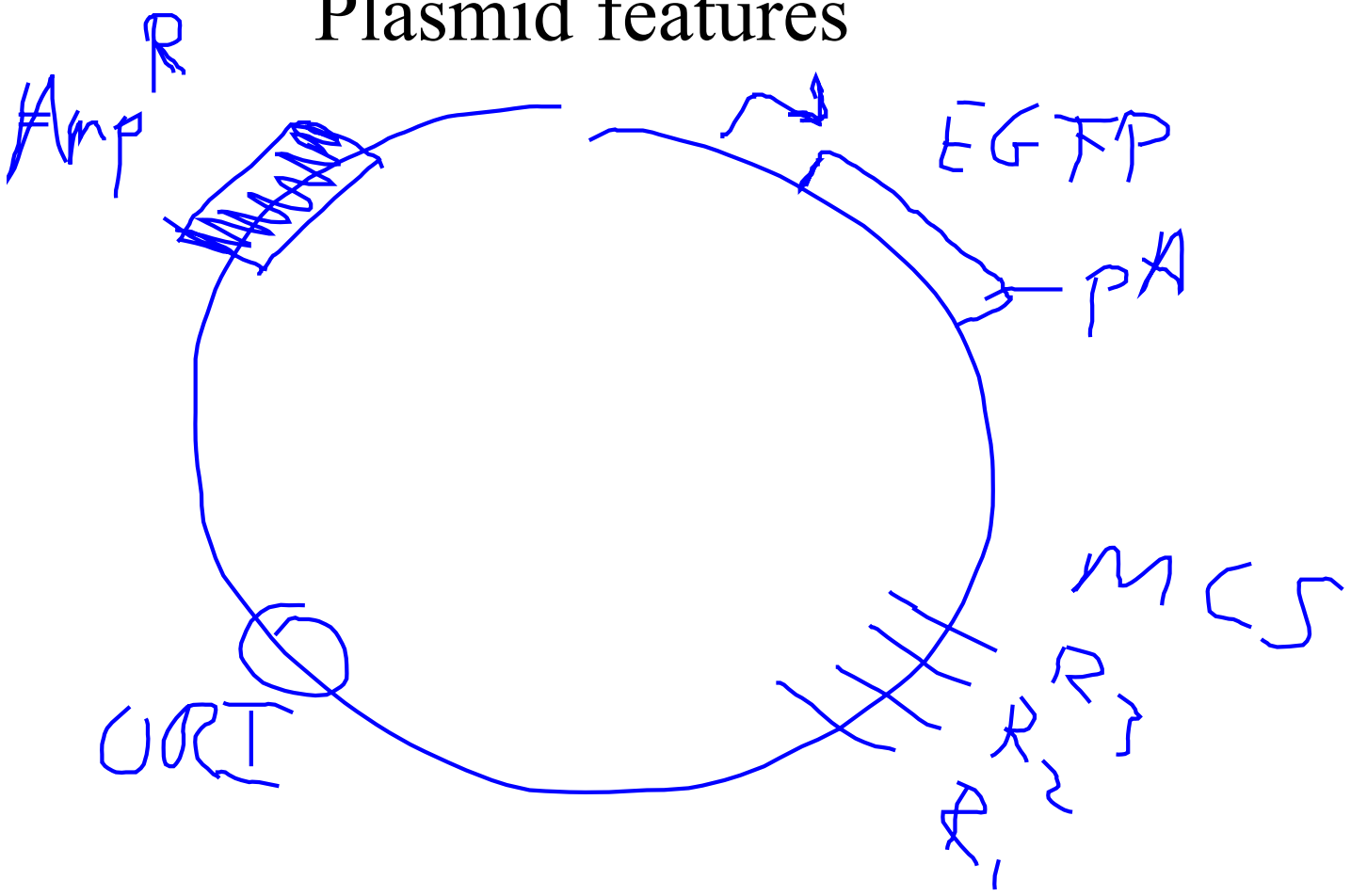
20.109 (F12)

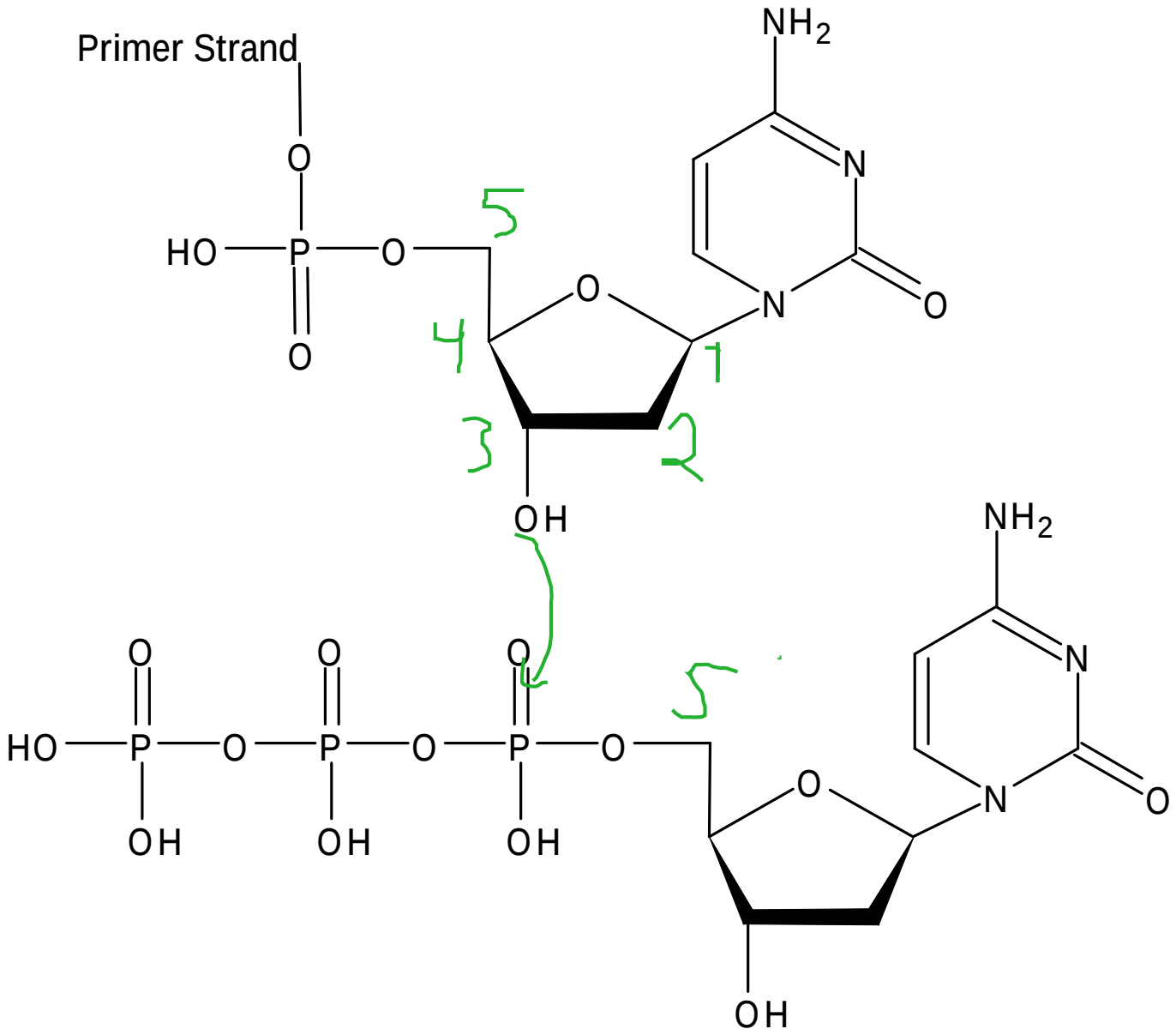
09.11.12

Module 1 overview

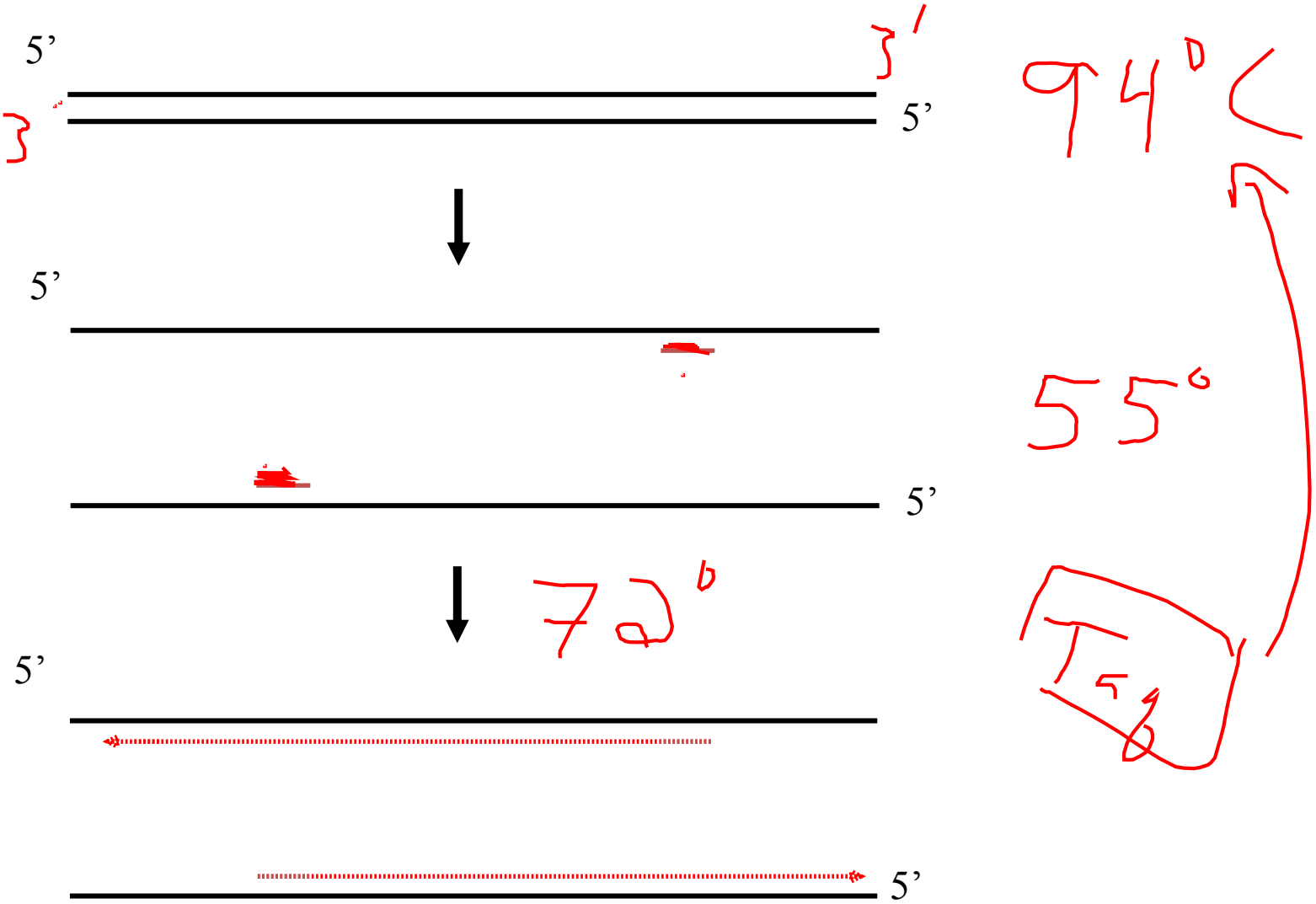


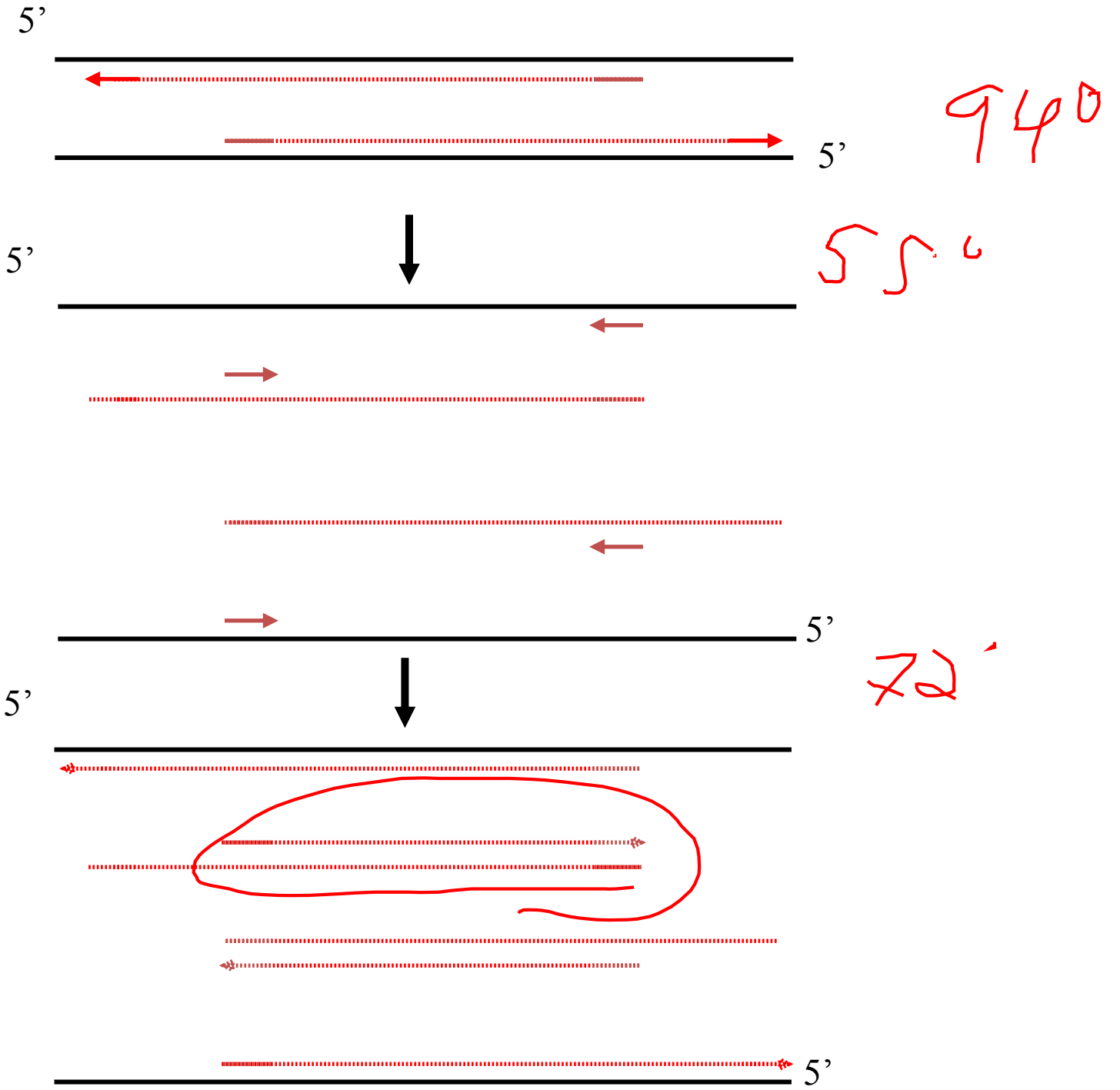
Plasmid features

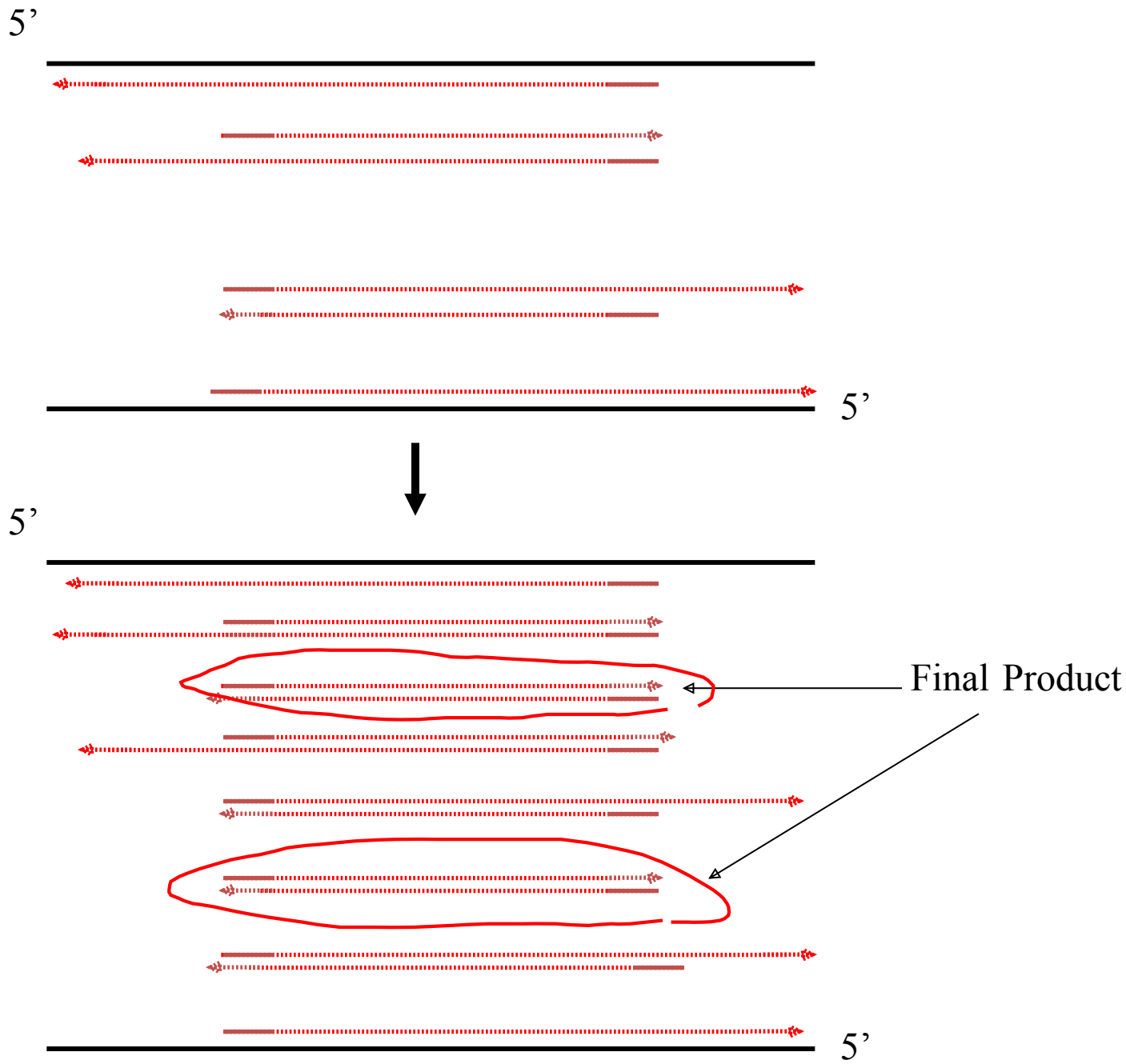



Primer Strand
NH2
O
HO
P
O
O
5
N
O
N
O
4
1
3
2
OH
NH2
O
O
O
HO
P
O
P
O
P
O
5
N
O
N
O
OH
OH
OH
OH

PCR

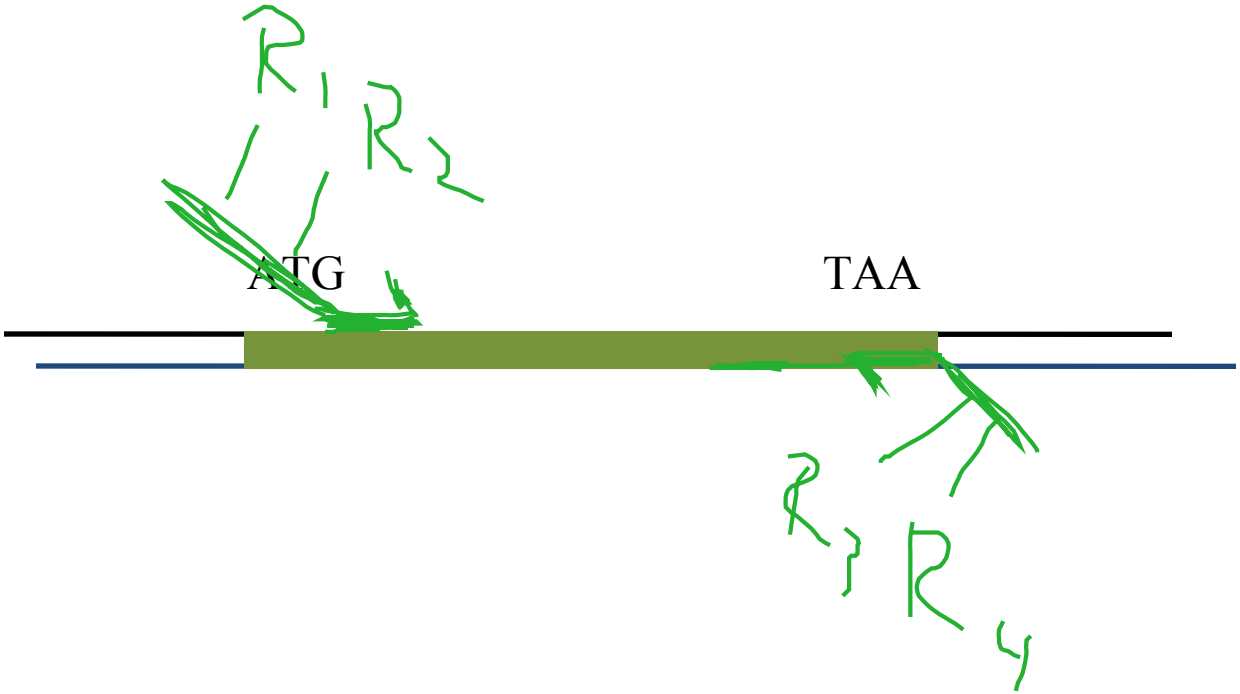



5’
5’
5’
5’
5’
5’
94°
55°
72°


5’
5’
5’
Final Product
5’

Your primers

delta 5:



Primer Design

1. 17-28 bases

2. 50-60% (GC)

3. Melting Temps should be ~65-80^{o}

4. 3'-ends of primers should not be complementary to each other (why?)

5. Hairpins should be avoided (why?)

6. Check for 'accidental' annealing elsewhere in your target.

Your reactions

Component	Function
PRIMERS	(2) Amplify / Define
TEMPLATE	source of seq
MASTER MIX	dNTPs
	Mg^{++} / Buffer
	Taq

20.109(F12):Guidelines for maintaining your lab notebook

20.109(F12): Laboratory Fundamentals of Biological Engineering

Evaluation [edit]

Grading your notebook [edit]

Once each module, the 20.109 teaching assistants will collect the duplicate copy of your notebook pages and evaluate them as follows:

Lab Notebook	Evaluation		
Date of experiment	√–	√	√+
Module#/Day#	√–	√	√+
Title for experiment	√–	√	√+
Brief statement of purpose	√–	√	√+
Protocol	√–	√	√+
Tables for data entry	√–	√	√+
Calculations entered	√–	√	√+
Data labeled	√–	√	√+
Summary/interpretation	√–	√	√+
Overall	√–	√	√+

OWW Basics

g

This week in lab

T: Lab certification
Primer Design
PCR

R: Clean up PCR product.
Digest DNA
Oral vs Written Comm.