Paternity Lab 3. Who's Your Daddy?

Objective

Using DNA sequence, determine paternity by using a restriction enzyme in several samples.

Background

Off the coast of Florida, at the Mote Marine Lab, Dr. Randy Wells and his colleagues are researching dolphin society, a task not easily accomplished. As they capture and release dolphins, they take certain samples in order to keep close records on how the dolphins are doing in the wild. A young dolphin calf that was swimming with a group of female dolphins has been brought onto Dr. Wells' research ship. He is aware of which female is the calf's mother but is unsure of the father's identity. He is also aware of which males frequent this area during the breeding season, and samples from them are stored. A blood sample, a swab of the calf's blowhole, and a tooth are taken from the calf before it is released. Dr. Wells brings the samples to you,

Dr. I. M. Yoo, for further analysis to figure out just who the father is.

Males who frequent this area during the breeding season:

Nemo (MML 1701)	Scotty (MML 1772)	Fetch (MML 1781)		
Stormy (MML 1802)	Plato (MML 1804)	Odie (MML 1811)		

- DNA is used by biologists for many purposes, such as identification. Since every individual's DNA is a unique combination of the maternal and paternal DNA, no other individual (except an identical twin) will look the same.
- Restriction enzymes are enzymes that cut DNA at a specific site. Once they make a cut, they continue down the DNA, cutting it each time they find their unique cutting site, which consists of a DNA sequence. For example, a restriction enzyme may look for the sequence AACCT and make a cut between the two Cs (AAC/CT).
- DNA is a polar molecule that is attracted to a positive charge.
- Gel electrophoresis is the process of separating DNA fragments by placing the fragments in a gel and running electricity through the gel. The shorter DNA segments move farther through the gel.
- A DNA fingerprint is the unique order of DNA segments that reflect each person's unique DNA. Fingerprints can be made through the process of gel electrophoresis.

Procedures

The steps involved in making a DNA fingerprint are as follows:

- 1. Isolate the DNA. This has been done for you. On the following page are several strands of each individual's DNA, along with DNA fragments that were taken from the mother and calf.
- 2. Prepare the DNA with restriction enzymes. You will simulate this by doing the following:
 - A. Read through each segment of DNA. Whenever you spot the sequence of bases CCTA, underline it, draw a slash after the T (example: CCT/A), and continue down the strand. You must start your counting over after each cut when moving down the sequence.
 - B. Each slash represents a spot where the restriction enzyme made a cut. Count and record how many bases are in each fragment of DNA. Each letter will be counted only once.
- 3. Separate the fragments with electrophoresis. You will simulate this by shading in the boxes in the Gel Electrophoresis data table. Shade in the boxes that correspond with the length of DNA fragments.



CalfDNA
Fragment Lengths
Nemo (MML 1701) GCGGATTTCCTACGATTTCGCCCCTATATATCGCGCGCCTAATAACCTACCT
Scotty (MML 1772) TATGGTTAGTTACCTACCCGGAACCTACCGCGCAATTAGCCCTAGGCCTAGGGCGTTATTCCTACATTTGCCTACTCCTAGCCACCTATACCTACC
Fragment Lengths
Fetch (MML 1781) GGTTCACACGGCCCTATCGGACCTAGCCATCATGCATCACCCTAGCCTAGCTAG
Stormy (MML 1802) GGAATTTCCTCGACCTAATACCTAGATATATCGCCTATCGCTCCCTAGTCGATATATCCTACACGCTCCTATATATGGGCTCACCTAATCGTATCGCTT Fragment Lengths
Plato (MML 1804) GGTTTAAATCGCTCCCCCTAATCGCCTAATCCCTATATCGCTAGGTCCTAATCCTAACCTAGATCCCCATCCTAATTAAT
Odie (MML 1811) CCTAGCTAGCTTCGAAATCACCTAGCCCTACATCCTAGTAATATATCCCCCTATCCCCCTAACGACTCCTAGATCAGCATCAGCATCCTATGGGCCCTC Fragment Lengths
Mother DNA TACCTACCCCAACGACCTAGTATCGACTCCTACCTCTACGATCAACCTACTCATCCCTAACGAACCTATACTAGCAGCCTAGTAGCTATCGGAT Fragment Lengths

Gel Electrophoresis

Wells	Calf DNA	Nemo MML1701	Scotty MML1772	Fetch MML1781	Stormy MML1802	Plato MML1804	Odie MML1811	Mother DNA
Fragment Lengths								
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Calf DNA

SOLUTION

Fragment Lengths 13/10/17/6/14/10/6/8/6/7 AATGCTTAGACCT / AGCGGATCCT / ACGGCGCTATTCGCCCT / ACGCCT / AGGGCGCTATTCCT / AGATTTCCCT / ACTCCT / AGCGACCT / AGACCT / AGCTAGC

Nemo (MML 1701)

Fragment Lengths 11/13/13/8/4/14/12/8/4/11 GCGGATTTCCT / ACGATTTCGCCCT / ATATATCGCGCCT / AATAACCT / ACCT / ATAGCTAGCATCCT / AGCCGCTATCCT / ACTTACCT / ACCT / AGTAGGCTAGA

Scotty (MML 1772)

Fragment Lengths 15/11/17/6/14/10/6/8/6/7 TATGGTTAGTTACCT / ACCCGGAACCT / ACCGCGCAATTAGCCCT / AGGCCT / AGGGCGTTATTCCT / ACATTTGCCT / ACTCCT / AGCCACCT / ATACCT / ACCTGGC

Fetch (MML 1781)

Fragment Lengths 15/9/19/5/11/13/9/5/11 GGTTCACACGGCCCT / ATCGGACCT / AGCCATCATGCATCACCCT / AGCCT / ACTCGCTACCT / AGATCACACACCT / AGTAGCCCT / AGCTCTCATGA

Stormy (MML 1802)

Fragment Lengths 16/7/13/10/14/10/16/13 GGAATTTCCTCGACCT / AATACCT / AGATATATCGCCT / ATCGCTCCCT / AGTCGATATATCCT / ACACGCTCCT / ATATATGGGCTCACCT / AATCGTATCGCTT

Plato (MML 1804)

Fragment Lengths 18/8/7/15/5/12/11/18 GGTTAAATCGCTCCCCCT / AATCGCCT / AATCGCCT / ATATCGCTAGGTCCT / ATCCT / AACCT / AGATCCCATCCT / AATTAATGCCT / AGCTGTATGCTAGCTTAA

Odie (MML 1811)

Fragment Lengths 3/20/6/7/15/7/10/19/10 CCT / AGCTAGCTTCGAAATCACCT / AGCCCT / ACATCCT / AGTAATATATCCCCT / ATCCCCT / AACGACTCCT / AGATCAGCATCAGCATCCT / ATGGGCCCTC

Mother DNA

Fragment Lengths 5/13/13/17/10/5/9/13/14 TACCT / ACCCCAACGACCT / AGTATCGACTCCT / ACCTCTACGATCAACCT / ACTCATCCCT / ACCCCT / AACGAACCT / ATACTAGCAGCCT / AGTAGCTATCGGAT

Gel Electrophoresis

SOLUTION

	Calf DNA	Nemo MML1701	Scotty MML1772	Fetch MML1781	Stormy MML1802	Plato MML1804	Odie MML1811	Mother DNA
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1								

Conclusions:

1) Compare the DNA fingerprints from the possible parents. Who is the father?

2) Explain your analysis in determining the paternity of the calf.

3) Is the entire set of DNA accounted for from both parents?	YES	NO	NOT ENOUGH INFO
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4) How would the paternity results change if a different restriction enzyme was used?

- 5) Why are genetic methods like this important for animals in zoos and in the wild?
- 6) The gel at the right is an actual paternity analysis of a family of dolphins. F is the father, M is the mother, and K1, K2, and K3 are the offspring. Circle the bands in the offspring that had to have come from the father and not the mother.

