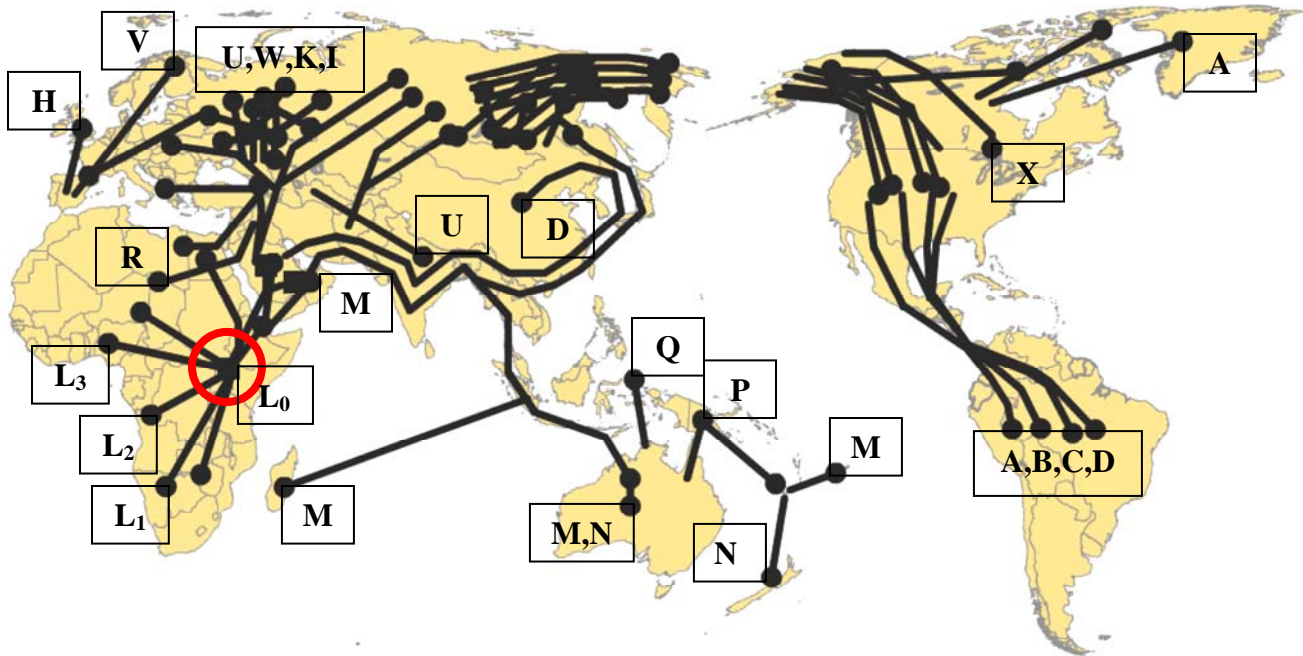


**A Human Migration Map**  
**Based on Mitochondrial DNA (mtDNA) Haplogroups**



*Letters on migration map correspond to mtDNA haplotypes*

**Human history told through the analysis of mitochondrial DNA (mtDNA)**

<b>Thousands of years ago</b>	<b>Event (Newly populated continent in brackets)</b>
200-160	Dawn of Mitochondrial Eve (Africa)
160-130	First steps outward
130-90	The peopling of Africa
90-70	First exit out of Africa (Asia)
70-65	In search of greener lands
65-55	Birth of "Adam" (Australia)
55-40	Trek across Asia
40-25	Expansion to other continents (Europe)
25-19	Bridging the gap - Beringia (North America)
19-15	In search of a refuge (South America)
15-12	The last ice age breaks
12-present	Colonization, domestication, agriculture

## How a Genetic Ancestry Kit Works

### Step 1

A swab of the inner cheek loosens cells that contain DNA.

### Step 2

DNA is extracted from either the mitochondria (to test matrilineage) or the nucleus (to test patrilineage or overall heritage).

### Step 3

The DNA is replicated, chopped into small pieces, split into single strands, and marked with a dye.

### Step 4

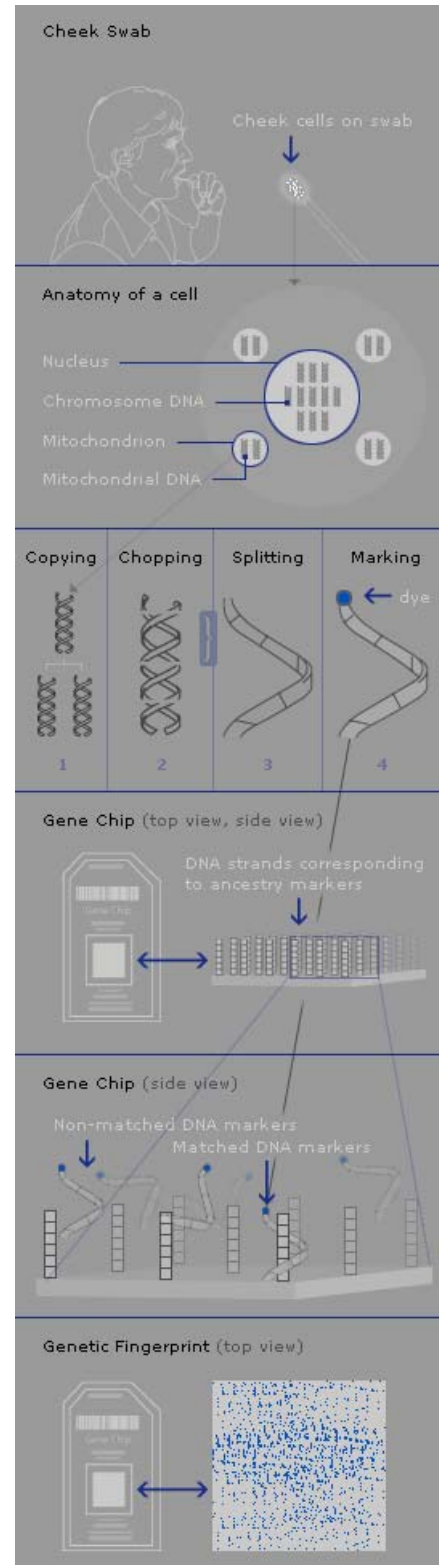
The chopped strands are then placed onto a gene chip that contains tens to hundreds of types of DNA corresponding to selected markers.

### Step 5

The strands only stick to spots on the chip that have matching markers.

### Step 6

Spots where the strands have stuck will glow, and the pattern of spots can be compared by computer to determine the person's genetic heritage.





## Tracing Human Ancestry with DNA Microarrays

### Instructions & Worksheet

#### **Background Information:**

You are a student genetic paleontologist on a dig of an ancient city, approximately 10,000 years old, in Eastern South America, what is known as Brazil today. You and your fellow student genetic paleontologists have been given the job of DNA testing any human remains that you find in order to help understand where the inhabitants migrated from, what their ancestry was, and to paint a fuller picture of the city, its inhabitants and their jobs or reasons for being in the city.

You have come across four different sites within the walls of this ancient city where you have found human remains entombed. You know from having studied about this ancient city and from what you can determine from the layout of the city that it was a Mecca of civilization. Having found walls and artifacts, the city appears to have had many streets with shops selling food, ceramics, and other household goods.

Remains of residences are quite abundant also. You can assume that the city was host to many different peoples who either lived there or traveled there.

Using DNA microarray technology, you will be able to determine the ancestry of the four humans you have sampled.

The results will help establish migration routes to the city from other parts of the world.

#### **Microarray Instructions and Worksheet (16 points)**

You will now be mimicking the DNA microarray process, with an on-paper assignment. The steps on this worksheet correspond to the steps on the page entitled "How a Genetic Ancestry Kit Works".

**Step 1 (Sample Collection):** Instead of taking a cheek swab as you would do in a home DNA kits, assume that you have collected soft tissue from one person buried at each of the four different sites.

**Step 2 (DNA Isolation):** The tissue samples contain cells that have both mitochondrial DNA and DNA from the Y-chromosome, but back in the lab, you can assume that you have isolated the mitochondrial DNA from the other cellular material.

**Step 3 (Replication, Chopping, Splitting, Marking):** The DNA has been replicated to give cDNA. The cDNA is then split into single strands, giving the four sets of cDNA in front of you. Cut the four sets of cDNA using scissors if they are not yet cut so that the strands are distributed between group members. Color the strands using a colored pencil or highlighter if they are not yet colored.

Now compare the cDNA strands. It will help to line up the four strands and use the edge of a ruler to see the similarities and differences.



## Tracing Human Ancestry with DNA Microarrays Instructions & Worksheet

a) How many of the bases are the same across all four strands? \_\_\_\_\_

b) How many of the bases are different? \_\_\_\_\_

Next, chop the strands using scissors as a "restriction enzyme." The restriction enzyme is a protein that cuts DNA in specific points so that DNA can be sequenced. For this experiment, cut the cDNA strands at every fourth base -- your cDNA pieces should each have four bases. Then mark the 5' end of each cDNA fragment with an asterisk (\*) signifying that the fragment has been tagged with a dye.

**Step 4 (The DNA Microarray):** Familiarize yourself with the DNA microarray master grid. This microarray has markers for five populations corresponding to mitochondrial DNA haplogroups. Microarrays can measure mutations as single nucleotide polymorphisms (SNPs) or microsatellites. This microarray examines SNPs found in mitochondrial DNA.

c) For the DNA on the microarray, which end points up, the 3' end or the 5' end? \_\_\_\_\_

**Step 5 (Hybridization):** Hybridize your chopped cDNA fragments to the complementary base sequences on the microarray grid master by attaching them with a glue stick or tape. You will need to remember or look up which bases hybridize.

Fill in the blank with the letter of the base that hybridizes with the bases listed below:

d) Adenine (A) : \_\_\_\_\_

e) Thymine (T) : \_\_\_\_\_

f) Guanine (G) : \_\_\_\_\_

g) Cytosine (C) : \_\_\_\_\_

h) How many of your fragments hybridized with the DNA microarray? \_\_\_\_\_

i) Are there any DNA fragments from your sample that did not hybridize to the Microarray Grid Master? \_\_\_\_\_

What is the most likely explanation for the non-hybridization of the fragment?

j) \_\_\_\_\_

\_\_\_\_\_



# EXTRA

## Tracing Human Ancestry with DNA Microarrays Instructions & Worksheet

**Step 6 (Analysis):** Once everyone on your team has attached all his/her fragments, take a look at the Microarray Master Grid. Look carefully at where the different colored fragments have been placed, then fill in the microarray analysis grid for your sample. This mimics the "readout" process of the DNA microarray.

*Determine which haplogroup (and location) each sample belongs to:*

k) Sample #1 Haplogroup: \_\_\_\_\_ Location: \_\_\_\_\_

l) Sample #2 Haplogroup: \_\_\_\_\_ Location: \_\_\_\_\_

m) Sample #3 Haplogroup: \_\_\_\_\_ Location: \_\_\_\_\_

n) Sample #4 Haplogroup: \_\_\_\_\_ Location: \_\_\_\_\_

*What conclusions can you make about the composition of the human population of this ancient city based on your analysis of the four different humans whose remains you analyzed?*

o) \_\_\_\_\_  
\_\_\_\_\_  
\_\_\_\_\_

Congratulations! You have determined genetic ancestry information by analyzing DNA with a microarray!



# EXTRA

## DNA SAMPLE STRANDS

#1  
(5' end)

T  
C  
C  
G  
A  
T  
A  
T  
C  
G  
T  
G  
A  
T  
C  
G  
C  
A  
G  
G  
A  
A  
C  
T  
G  
G  
T  
C  
T  
T  
G  
A  
T  
T  
A  
C  
T  
T  
T  
T

(3' end)

#2  
(5' end)

T  
A  
G  
C  
A  
T  
A  
T  
C  
G  
T  
G  
A  
T  
C  
G  
C  
C  
C  
A  
A  
C  
G  
T  
G  
A  
T  
T  
T  
T  
G  
A  
T  
T  
A  
C  
T  
G  
A  
T

(3' end)

#3  
(5' end)

T  
C  
T  
A  
A  
T  
A  
T  
C  
G  
T  
G  
A  
T  
C  
G  
C  
T  
T  
A  
A  
T  
T  
C  
C  
T  
G  
T  
T  
T  
G  
A  
T  
T  
A  
C  
C  
C  
G  
A  
A

(3' end)

#4  
(5' end)

T  
T  
A  
A  
A  
T  
A  
T  
C  
G  
T  
G  
A  
T  
C  
G  
C  
A  
C  
C  
A  
G  
G  
G  
G  
A  
A  
G  
T  
T  
G  
A  
T  
T  
A  
C  
G  
T  
G  
T

(3' end)



## Microarray Master Grid

A A A A	A A T T	A C T A	A G A T	A G G C
A T C G	C A C A	C A C A	C A T G	C C A G
C T A A	C T T C	G A A T	G A C A	G C T T
G G G T	G G T T	G T C C	G T G G	G T T A
T A A G	T C C C	T G C A	T T G A	T T T G



## Microarray Analysis Grids


**Sample #1**


**Sample #2**


**Sample #3**


**Sample #4**

		*	*	
	*			*
				*

**Haplogroup Q  
(Indonesia)**

		*		
*				
*				
*				
		*		

**Haplogroup A  
(South America)**

			*	
		*	*	*
*				

**Haplogroup L<sub>1</sub>  
(Eastern Africa)**

*				*
				*
		*		
			*	

**Haplogroup U  
(Eastern Europe)**

	*			
	*			
	*			
			*	
	*			

**Haplogroup V  
(Scandinavia)**



## Tracing Human Ancestry with DNA Microarrays

### **Homework Essay:**

Write a one-page essay on the ethical issues surrounding genetic ancestry tests. Be ready to discuss your thoughts in the next class. Use your answers to one or more of these questions to help you develop your essay.

1. Can genetics be used to define race?
2. How much faith can you put in a result from a DNA ancestry test?
3. What are some ethical issues associated with profit-making companies collecting genetic information?
4. Can potential harm caused by genetic testing be balanced by potential good done when applied the tests for medical research?



## Tracing Human Ancestry with DNA Microarrays

### Homework Questions (16 points):

The following Web sites are excellent descriptive and demonstrative examples of DNA microarray technology:

The Online NewsHour's in-depth coverage of genetic ancestry, available at [http://www.pbs.org/newshour/indepth\\_coverage/science/dna/](http://www.pbs.org/newshour/indepth_coverage/science/dna/) and "DNA Kits Provide Insight into Genetic Ancestry", available at [http://www.pbs.org/newshour/bb/science/july-dec06/ancestry\\_07-20.html](http://www.pbs.org/newshour/bb/science/july-dec06/ancestry_07-20.html)

More information about microarrays:

<http://www.hhmi.org/biointeractive/genomics/microarray.html> (Howard Hughes Medical Institute)

<http://www.ncbi.nlm.nih.gov/About/primer/microarrays.html> (National Center for Biotechnology Education)

<http://affymetrix.com/corporate/outreach/educator.affx> (Affymetrix)

<http://www.bradenton.com/mld/bradenton/news/local/15256754.htm> (Herald Today / Associated Press)

<http://www.bio.davidson.edu/courses/genomics/chip/chip.html> (Davidson)

<http://gslc.genetics.utah.edu/units/biotech/microarray> (Genetic Science Learning Center, University of Utah)

Using the above links, answer these questions:

1. How are the various types of cells in our bodies different from one another, genetically speaking?
2. What does 'gene expression' mean?
3. Why do researchers generally purchase already manufactured microarrays from biotechnology companies rather than make them in their own labs?
4. Each spot on the microarray corresponds to what?
5. A DNA sequence of C-A-T-T-G will stick to, or hybridize to the following base sequence \_\_\_\_\_ to form double-stranded DNA.
6. DNA sequence arrays can be used to detect what?
7. There are two great values of using microarray technology. What are they?
8. What is the biological source of restriction enzymes?



## Tracing Human Ancestry with DNA Microarrays

### Worksheet Answers

- a) How many of the bases are the same across all four strands? 23
- b) How many of the bases are different? 17
- c) For the DNA on the microarray, which end points up, the 3' end or the 5' end? 3'

Fill in the blank with the letter of the base that hybridizes with the bases listed below:

- d) Adenine (A) : T
- e) Thymine (T) : A
- f) Guanine (G) : C
- g) Cytosine (C) : G
- h) How many of your fragments hybridized with the DNA microarray? 5
- i) Are there any DNA fragments from your sample that did not hybridize to the Microarray Grid Master? YES (5)

What is the most likely explanation for the non-hybridization of fragments?

- j) The DNA markers are common to two or more haplogroups so they aren't included in the section of the microarray for unique markers. It could be a gene that codes for a protein that is found in all human beings, perhaps the Cytochrome C protein.

Determine which haplogroup (and location) each sample belongs to:

- k) Sample #1 Haplogroup: U Location: Eastern Europe
- l) Sample #2 Haplogroup: A Location: South America
- m) Sample #3 Haplogroup: L<sub>1</sub> Location: Eastern Africa
- n) Sample #4 Haplogroup: V Location: Scandinavia

Note: Haplogroup Q (Indonesia) does not match with any sample.

What conclusions can you make about the composition of the human population of this ancient city based on your analysis of the four different humans whose remains you analyzed?

- o) Certainly, the population was diverse with individuals having come there as travelers from various parts of the world.



## Tracing Human Ancestry with DNA Microarrays

### Homework Answers

1. All body cells contain the same DNA. The difference is in which genes are 'expressed' or turned on in the particular cells. For instance, the genes that code for actin and myosin, expressed in muscle cells, would not be expressed in skin cells.
2. Gene expression relates to the function of the gene. If the gene is 'expressed' in a cell, then it is producing a protein. If the gene is not expressed, then that protein is not being made in that cell.
3. It takes many steps, many different pieces of equipment, and thousands of DNA samples to make a microarray. Because of the time and energy a lab would need to manufacture its own microarrays, a lab generally chooses to spend money on a pre-manufactured microarray.
4. Each spot corresponds to one gene, and contains many probes, or the sequence for that one gene.
5. G – T – A – A – C
6. Microarrays can be used to detect SNPs, which are single nucleotide polymorphisms, usually harmless genetic mutations which accumulate over generations in the genomes of living organisms.
7. A scientist can analyze a lot of DNA at one time. And he/she can compare the DNA from two sources at the same time by computer analyzing the pattern of colors caused by the fluorescent tags on the DNA.
8. Restriction enzymes come from bacteria. They are thought to be defense mechanisms to aid the bacteria in destroying their enemies by cutting the enemy DNA into fragments.