

## Adaptation by Natural Selection

## Natural Selection

- Darwinian natural selection is a two-step process:
  - The production of new genetic variation through the process of mutation
  - The differential reproduction of favorable variants through the process of selection

## Selection

- Selection causes changes in allele and genotype frequencies from one generation to the next due to differential net reproductive success of individuals with different genotypes
  - If individuals with genotype AA consistently have twice as many offspring as individuals with AB and BB genotypes, the frequency of the A allele will increase through time and eventually, everyone will have the AA genotype

## Selection, 2

- There are two elements contributing to the differential reproductive success of individuals with differing genotypes
  - Viability or survival: individuals must survive to reproductive maturity in order to be able to reproduce
  - Fertility: individuals must produce offspring in order to pass on their genes on to the next generation of the population

## Modeling Selection

- Selection operates by reducing the completed fertility of individuals with a certain **phenotype**, relative to other phenotypes within a population
  - Fitness ( $1.0 \leq w \leq 0.0$ ) is the completed fertility for a given genotype, relative to the genotype with the highest completed fertility
  - The selection coefficient ( $1.0 \leq s \leq 0.0$ ) measures the relative reduction in fertility for a genotype
    - $s = 1 - w$

## Modes of Somatic Selection

Autosomal Inheritance	Genotypes		
Selection Against:	AA	AB	BB
Complete Dominance			
Dominant (Huntington's Disease)	S <sub>AA</sub>	S <sub>AA</sub>	
Recessive (PKU, Tay Sachs)			S <sub>BB</sub>
Incomplete Dominance/Codominance			
Heterozygote		S <sub>AB</sub>	
One homozygote, heterozygote	S <sub>AA</sub>	S <sub>AB</sub>	
Both homozygotes (sickle cell)	S <sub>AA</sub>		S <sub>BB</sub>

## Modes of Sex-linked Selection

Traits on X Chromosome	Females			Males	
Selection Against:	AA	AB	BB	A	B
Complete Dominance					
Dominant	$s_{AA}$	$s_{AA}$		$s_{AA}$	
Recessive (Hemophilia A)			$s_{BB}$		$s_{BB}$
Incomplete Dominance					
Heterozygote		$s_{AB}$			
1 homozygote, heterozygote	$s_{AA}$	$s_{AB}$		$s_{AA}$	
Both homozygotes (G6PD)	$s_{AA}$		$s_{BB}$	$s_{AA}$	$s_{BB}$
Traits on Y Chromosome					
Any allele (A)				$s_{AA}$	

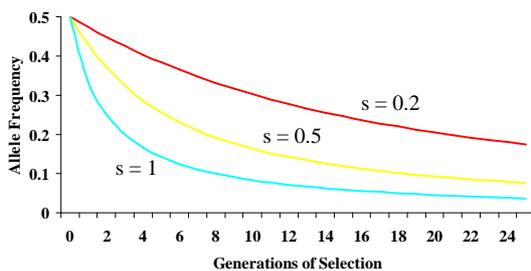
## Selection against a recessive

	AA	AB	BB
Hardy-Weinberg Freq:	$p^2$	$2pq$	$q^2$
Fitness (w)	1.0	1.0	$1 - s_{(BB)}$
Frequency after selection	$p^2 \times (1)$	$2pq \times (1)$	$q^2 \times (1-s)$
New Total Frequency ( $\bar{w}$ ) = $p^2 + 2pq + q^2 - q^2s = 1 - q^2s$			
Relative frequency after selection	$\frac{p^2}{(1 - q^2s)}$	$\frac{2pq}{(1 - q^2s)}$	$\frac{q^2 - q^2s}{(1 - q^2s)}$

Frequency of B in next generation = freq (BB) + ½ freq (AB)

$$q_1 = \frac{(q - q^2s)}{(1 - q^2s)}$$

## Selection against a recessive



## Overdominance

	AA	AB	BB
Hardy-Weinberg Freq:	$p^2$	$2pq$	$q^2$
Fitness (w)	$1 - s_{AA}$	1.0	$1 - s_{BB}$
Frequency after selection	$p^2 \times (1 - s_{AA})$	$2pq \times (1)$	$q^2 \times (1 - s_{BB})$
New Total Frequency ( $\bar{w}$ ) = $1 - p^2s_{AA} - q^2s_{BB}$			
Relative frequency after selection	$\frac{(p^2 - p^2s_{AA})}{(\bar{w})}$	$\frac{(2pq)}{(\bar{w})}$	$\frac{(q^2 - q^2s_{BB})}{(\bar{w})}$

Frequency of B in next generation = freq (BB) + ½ freq (AB)

$$q_1 = (q - q^2s_{BB}) \div (1 - p^2s_{AA} - q^2s_{BB})$$

Equilibrium is attained if  $\Delta q = 0$ ;

$$\Delta q = q_1 - q_0 = [(q - q^2s_{BB}) \div (1 - p^2s_{AA} - q^2s_{BB})] - q$$

$$= [pq \times (ps_{AA} - qs_{BB})] \div (1 - p^2s_{AA} - q^2s_{BB})$$

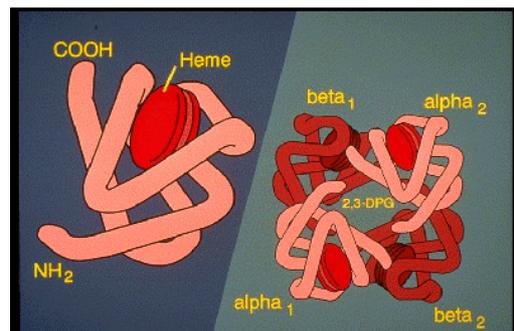
$$\Delta q = 0 \text{ if } ps_{AA} - qs_{BB} = 0; \text{ that is, if } q = \frac{s_{AA}}{(s_{AA} + s_{BB})}$$

## Sickle Cell Hemoglobin and Malaria

Natural Selection in Human Populations

## Structure of Hemoglobin

- 4 polypeptides (globins) and 4 iron-based oxygen-binding heme molecules



## Genetics of Hemoglobin

- The alpha and beta globin genes are transmitted separately, as parts of the 16th and 11th chromosome pair, respectively
  - Transmission follows Mendel’s Law of Independent Assortment for genes located on different homologous chromosome pairs
  - Each individual has two alpha globin genes and two beta globin genes inheriting one gene of each pair from the mother and one from father

## Function of Hemoglobin

- Hemoglobin is the primary protein constituent of red blood cells
  - Transports oxygen by binding with it tightly as the red blood cells pass through the capillaries of the lungs
  - As oxygenated red blood cells circulate through the heart and to the other body tissues, hemoglobin loosens its hold on the oxygen so that it can pass readily out of the red cells and be made available to peripheral cells for respiration

## Function of Hemoglobin 2

- Deoxygenated hemoglobin molecules in peripheral capillaries bind loosely with carbon dioxide and help remove this waste product of cellular respiration to the lungs for expiration
- The binding capacity of hemoglobin molecules is a function of the partial pressure of oxygen in the blood
  - high in the lungs, bind tightly
  - low in peripheral capillaries, bind loosely

## Hb<sup>A</sup>

- “Wild” or most common form, found in all human populations

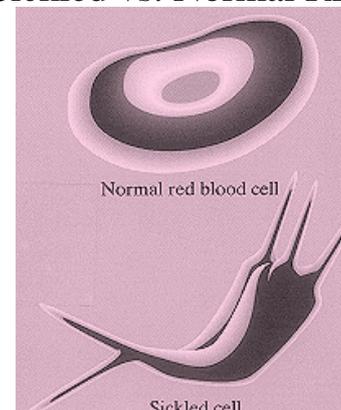
Beta Hemoglobin	6 <sup>th</sup>	26 <sup>th</sup>
DNA sequence	C-T-C	C-T-C
Amino Acids	Glutamic Acid	Glutamic Acid

## Hb<sup>S</sup>

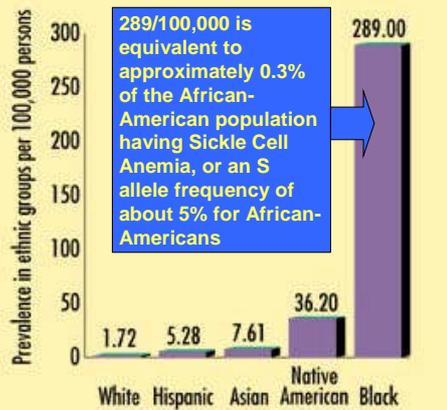
- Sub-Saharan Africa, Mediterranean, Middle East, South Asia

Beta Hemoglobin	6 <sup>th</sup>	26 <sup>th</sup>
DNA sequence	C-A-C	C-T-C
Amino Acids	Valine	Glutamic Acid

## Sickled vs. Normal RBC



### Sickle Cell Disease in the United States



### Hb<sup>C</sup>

- Predominantly West African populations
- |                 |                 |                  |
|-----------------|-----------------|------------------|
| Beta Hemoglobin | 6 <sup>th</sup> | 26 <sup>th</sup> |
| DNA sequence    | T-T-C           | C-T-C            |
| Amino Acids     | Lysine          | Glutamic Acid    |

### Hb<sup>E</sup>

- Southeast Asia
- |                 |                 |                  |
|-----------------|-----------------|------------------|
| Beta Hemoglobin | 6 <sup>th</sup> | 26 <sup>th</sup> |
| DNA sequence    | C-T-C           | T-T-C            |
| Amino Acids     | Glutamic Acid   | Lysine           |

### Genetics of Sickle Cell

- Three genotypes can form from combinations of the A and S alleles
  - AA Homozygous Normal; “Normal”
  - AS Heterozygote; “Sickle Cell Trait”
  - SS Homozygous sickler; “Sickle Cell Anemia”
- Inheritance follows a Mendelian pattern for a co-dominant autosomal allele
  - When two heterozygotes mate, ¼ of their offspring are predicted to be SS and have sickle cell anemia, ¼ AA normal, and ½ AS, carrying the sickler allele

### Sickling

- Red blood cells begin to sickle when hemoglobin molecules have given up their oxygen in the capillaries
- The S-hemoglobin molecules bind together into long fibers, forming a complex helical molecule within the red blood cell

### Sickling 2

- The formation of these fibers is dependent on the presence of valine at the β-6 position to bind one beta chain with the beta chain of another hemoglobin molecule
- For sickling of the red blood cell to occur it takes from 0.1 to 1 second of extreme deoxygenation of the hemoglobin molecules

### Sickling 3

- The deoxygenation necessary for sickling occurs in the capillaries after oxygen has been given off to the muscle cells and before the RBCs can return to the lungs
- Cells with higher concentrations of hemoglobin S form fibers more rapidly and are therefore more likely to sickle

### Sickling 4

- Hemoglobin S concentration is a function of cell age, with the concentration increasing as the red blood cell ages
- Hence, the older the red blood cells, the more likely they are to sickle
- The result is a downward spiral of continuous depletion of red blood cells, resulting in anemia

### Symptoms of Sickle Cell Anemia

- As a result of sickling and the premature aging of red blood cells from sickling, there are fewer than normal red blood cells, the general condition referred to as anemia
- There is an increased risk of severe infections, especially bacterial infections--such as sepsis (a blood stream infection), meningitis, and pneumonia, especially in early childhood
  - The risk of infection is increased because the spleen does not function normally

### Symptoms 2

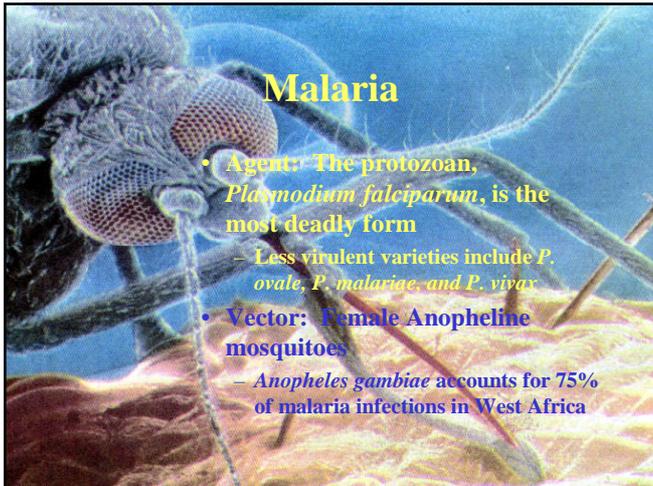
- Splenic sequestration crisis:
  - The spleen is the organ that filters blood
  - In children with sickle cell disease, the spleen can enlarge rapidly from trapped red blood cells creating a situation that can be life-threatening.
- Stroke:
  - This happens when blood vessels in the brain are blocked by sickled red blood cells
  - Signs include seizure, weakness of the arms and legs, speech problems, and loss of consciousness.

### Symptoms 3

- Children with sickle cell anemia experience slowed growth and delayed maturation, including puberty as a result of the anemia and infections
- There are repeated, painful episodes, called vaso-occlusive crises, associated with blockages of the circulatory system
  - Frequently seen as swelling of extremities
- There is a progressive degeneration of organs from impaired circulation

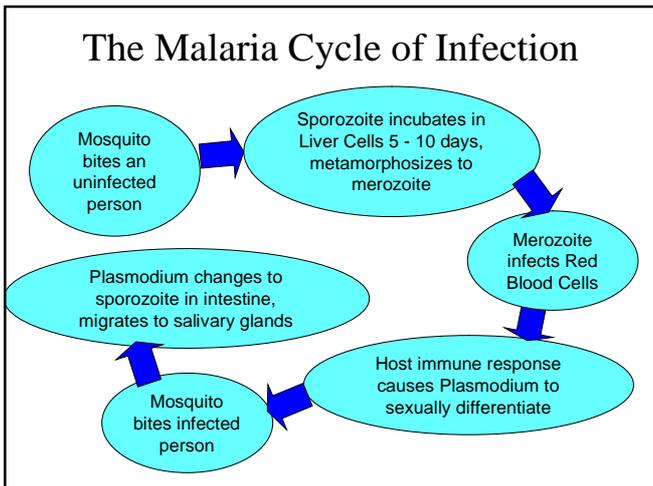
### Swollen Hands from Sickling





## Malaria

- **Host:** Man, although higher primates can also may harbor *P. malariae*
  - Monkeys harbor other plasmodium species which can infect man
- **Symptoms:** Cyclic (sometimes) high fever, chills and sweating, headache, coagulation defects, shock, anemia (inducing jaundice), kidney failure, acute encephalitis, coma

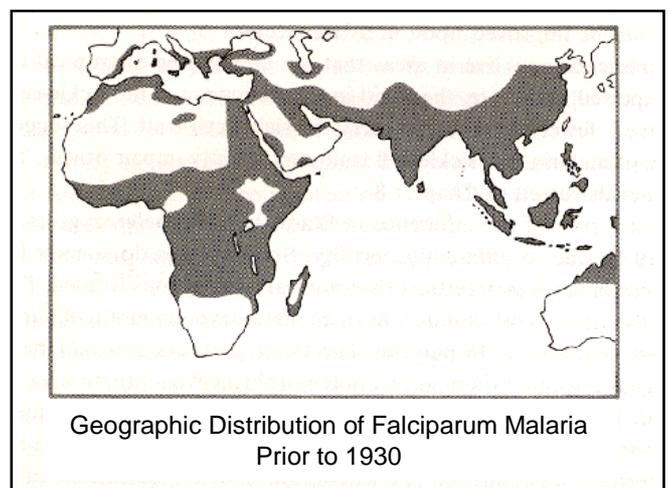


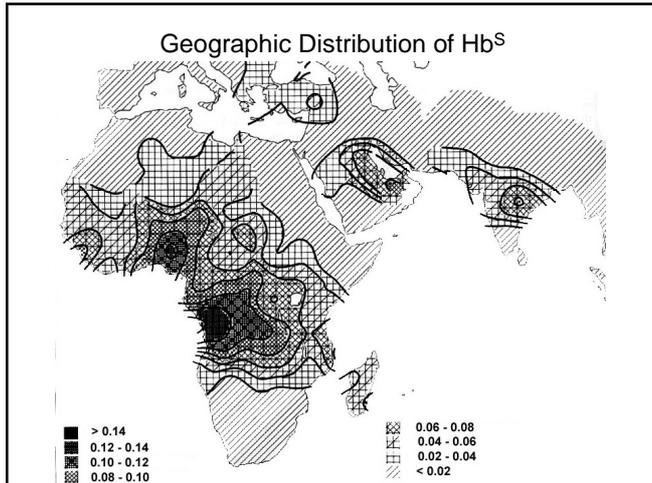
## Sickle Cell and Malaria

- There are three lines of evidence suggesting an association between the sickle cell allele and Malaria:
  - Geographic correlations
  - Epidemiological associations
  - Biochemical studies

## Geographic Correlations

- Clinical studies show a substantial overlap between the distribution of malaria in the and the frequency of the sickle cell allele
  - The Malaria belt extends across the Southern Mediterranean, sub-Saharan Africa, the Middle East, India, Southeast and Island Southeast Asia, and Northern Australia, the S allele exceeds a frequency of 10% only in Africa
  - In the Mediterranean  $\beta$  thalassemia and G-6-PD are elevated, while in India and Southeast Asia, Hemoglobin E and  $\alpha$  thalassemia predominate





## Epidemiological Correlations

- A comparison of *Plasmodium falciparum* parasites in blood samples from children of AA and AS genotypes in Nigeria to that of children with AA genotypes showed:
  - AS children had lower frequencies and lower densities of parasites than AA children
  - Fertility did not differ between AA and AS, but 29% more AS individuals survived to adulthood

## Biochemical studies

- *Plasmodium* metabolism causes sickle cell hemoglobin to form the fibers that results in red blood cell sickling
  - The parasite significantly lowers cytoplasm pH (increases acidity) causing hemoglobin to release oxygen
  - “Knobs” form on the cell wall of infected RBCs slowing transit through capillaries
  - The result is an increase of sickling to about 40% of the RBCs in AS individuals

## Biochemical studies 2

- The development of the *Plasmodium* is disrupted by the sickling of the red blood cells
  - Sickling depletes cell reserves of potassium which is required for the parasite to grow
    - *Plasmodium* quickly dies in potassium depleted cells
  - Additionally, infected sickled cells may be prematurely removed by the spleen

## Biochemical studies 3

- Sickling of a significant proportion of infected cells increases malaria resistance, even if not all parasites die
  - The subsequent reduction in the rate of multiplication of *Plasmodium* gives the immune system time to mount a serious counterattack

Why does sickle cell reach such high frequencies in West Africa?

## Cultural Factors

- Spread of swidden agriculture into previous Tropical Rainforest regions 2-3,000 years ago
  - The invasion of rainforest areas is facilitated particularly by the arrival of the Malaysian Crop Complex from Indonesia
    - Carried by sailors from Borneo across the Indian Ocean, contact established with Madagascar (native language is an Indonesian variety), then spread across forest and woodland areas of continental Africa

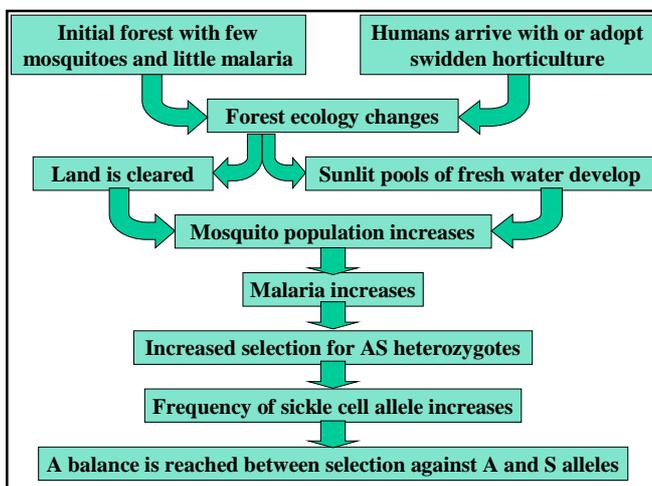
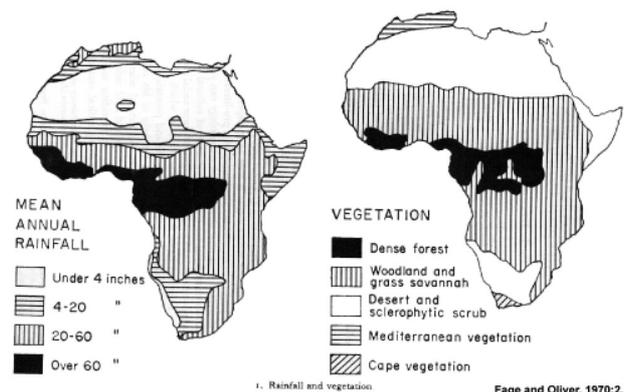
## Cultural Factors, 2

- As the root (*Dioscorea* yams) and tree (*Musa* bananas and plantains) crops spread, increasing amounts of tropical rainforest are cleared, supporting larger and larger human populations
- Clearing of the forest and high levels of rainfall erodes the soils which are lateritic and relatively impermeable, making for formation of pools of water

## Cultural Factors, 3

- During the rainy season pools of fresh water form, exposed to sunlight because of the forest clearing, which along with high humidity creates an ideal breeding habitat for *Anopheles gambiae*
  - Best predictor of malaria infection is various measures of rainfall, excess water, relative humidity
- Agricultural expansion supports more people, living in sedentary villages with huts that the mosquitoes use

## Rainfall and Vegetation of Africa



## A Yoruba Example

Genotype	Number of Adults (obs.)	Expected Frequency	Expected Number of Adults	Observed / Expected	Relative Fitness	Selection Coefficient
Hb <sup>A</sup> Hb <sup>A</sup>	9,365	0.769	9,525.6	0.98	0.88	0.12
Hb <sup>A</sup> Hb <sup>S</sup>	2,993	0.216	2,675.6	1.12	1.00	0.00
Hb <sup>S</sup> Hb <sup>S</sup>	29	0.015	185.8	0.16	0.14	0.86
Total	12,387	1.000	12,387			

The difference in selection coefficients between the the three genotypes is based on the difference in mortality between the three groups

Numerous studies have examined differential fertility and mortality based on Hb genotype and they have found most commonly that there are no fertility differentials, only mortality differentials

### Diet and Sickle Cell

- Today the consumption of cassava (*Manihot esculenta*) may confer an adaptation to both malaria and sickling and thereby disrupt the prehistoric balance of selective pressures
- Cassava is a significant source of biologically active cyanide compounds including cyanide (CN<sup>-</sup>), thiocyanate (SCN<sup>-</sup>), and cyanate (CNO<sup>-</sup>)

### Diet and Sickle Cell, 2

- At sublethal levels these compounds carbamylate polypeptides such as  $\beta$ -globin, Glucose-6-phosphate dehydrogenase, and other protein constituents by binding with the amine (NH) end of the peptide chain
  - This carbamylation prevents the formation of the sickle threads in deoxygenated sickle cells
  - Enhances likelihood of survival of individuals with sickle cell anemia but reduces the malarial resistance of heterozygotes

### Diet and Sickle Cell, 3

- Carbamylation of Glucose-6-phosphate dehydrogenase reduces the activity of that enzyme, emulating G-6-PD deficiency to the extent that the enzyme is carbamylated
- High circulating levels of cyanide metabolites may directly interact with *Plasmodium falciparum* to disrupt growth and reproduction of the parasite inside red blood cells
  - Lower levels carbamylate sickle cell hemoglobin

### Diet and Sickle Cell, 4

- Cassava cannot have played a role in the evolution of sickle cell frequencies since it was introduced as a significant cultigen in West Africa in 1620
- But Yams yield smaller amounts of thiocyanate which may have played a role historically in selection related to malaria and sickle cell hemoglobin

### Malaria, Sickle Cell, and Cyanates

	AA	AS	SS
Fertility	Normal	Normal	Low
Mortality	High	Low	Very high
Mortality add dietary cyanates	Moderate	Low	High

### Geographic Distribution in Africa

- There are several centers of high frequency of the S allele in Central Africa and West Africa
  - DNA sequencing of the non-coding areas of the Beta-globin locus show different sequences on the mutated 16th chromosome of the Hb<sup>S</sup> carriers in the different regions

### Sickle Cell Mutations

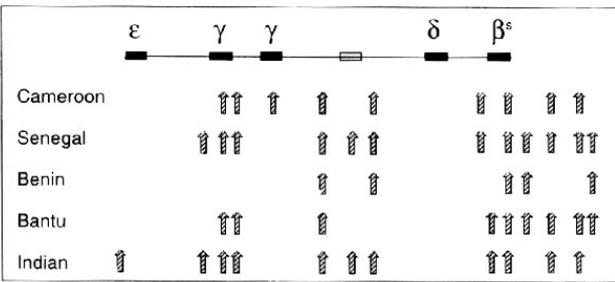
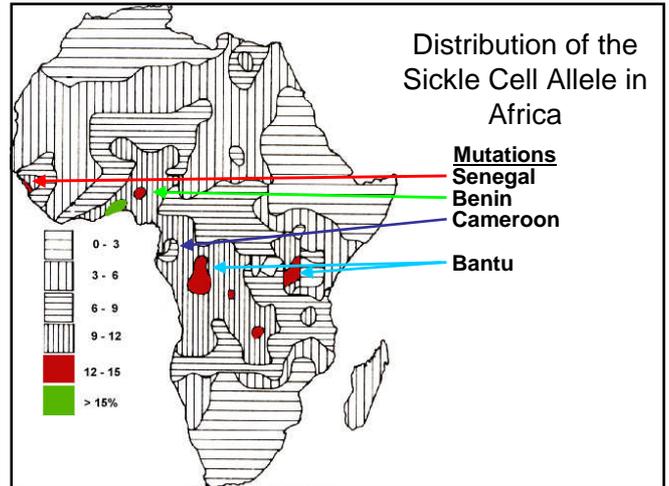


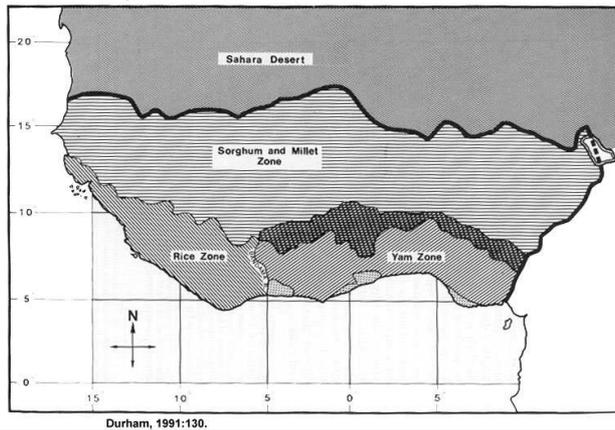
Figure 8.7. A map of restriction sites shows that the sickle cell mutation has arisen several times in several genomic surroundings or haplotypes. Each is named for a geographical area where it is common. Marks, 1995:147.



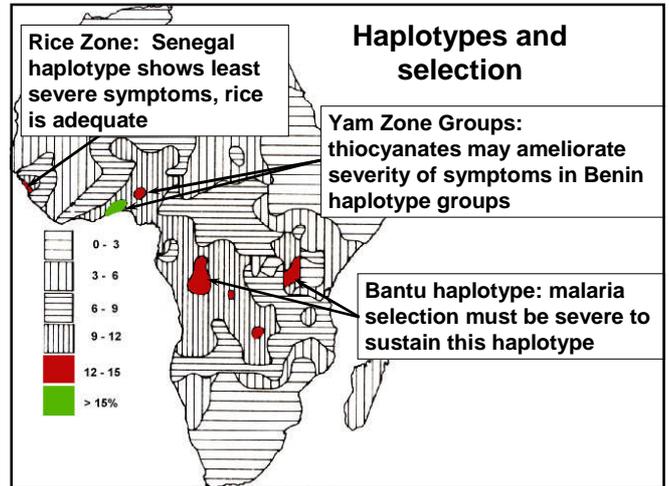
### Distribution of the Sickle Cell Allele in Africa

Mutations  
Senegal  
Benin  
Cameroon  
Bantu

### Agricultural Zones of West Africa



Durham, 1991:130.



Rice Zone: Senegal haplotype shows least severe symptoms, rice is adequate

### Haplotypes and selection

Yam Zone Groups: thiocyanates may ameliorate severity of symptoms in Benin haplotype groups

Bantu haplotype: malaria selection must be severe to sustain this haplotype

### Nitric Oxide

- Recent work has demonstrated that the pain associated with sickle cell crises is linked to excess hemoglobin in the blood
  - Pain has been described as akin to child birth or terminal cancer

### Sickle Cell Crisis

- When RBCs sickle membrane is destroyed releasing contents into the blood stream
  - The bulk of the contents is hemoglobin
  - Hemoglobin in the blood has high affinity for nitric oxide (NO)
    - Hemoglobin in blood binds 1000X greater than when in RBCs
  - Free hemoglobin scours nitric oxide
    - NO is a short-lived gas produced by cells lining blood vessels
      - Serves to regulate blood flow: prevents aggregation of platelets (clotting), protects cell membranes, relaxes vessels

## Sickle Cell and Nitric Oxide

- Blood vessels constrict and become clot prone as a result of hemoglobin reducing NO in the circulatory system
  - Increases blood pressure in lungs, restrict flow of oxygen, nutrients to vital tissues and organs
  - Severe pain ensues
- Potential for treatment for sickling crises
  - Breathe higher doses of nitric oxide to neutralize hemoglobin effect
  - Administer supplemental supplies of haptoglobin, the protein in our blood which normally scavenges hemoglobin

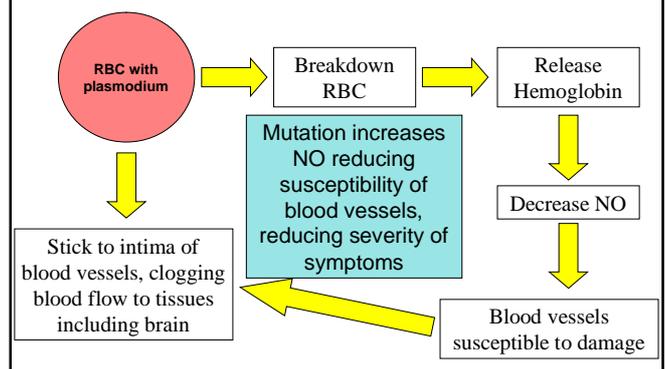
## A “New” Genetic Adaptation to Malaria

- Nitric oxide inhibits the growth of *Plasmodium in vitro*
- Researchers searched for a variant of the gene that regulates NO
  - NOS2: Nitric Oxide Synthetase 2, (chromosome 17 q arm near centromere)
  - Examined 1291 children in Tanzania and Kenya
    - Those with a specific mutation of NOS2 were much less likely to get severe malaria

## NOS2

- Risk reduction of severe malaria was 88% for the Tanzanian children and 75% for the Kenyan children
  - Two different varieties of malaria involved
- Children with the mutation produce greater amounts of NO

## Nitric Oxide and Malaria



## More Nitric Oxide and Malaria

- Alteration of the Anopheles gene analogue to human NOS2 affects the ability of the mosquito to transmit malaria
  - Plasmodium life cycle in the mosquito is interrupted by increased NO