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Substructuring in four populations of African descent

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SUBSTRUCTURING IN FOUR POPULATIONS OF AFRICAN DESCENT

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by

Margarita Teresa Almeida

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To: Dean Arthur W. Herriott
College of Arts and Science

This thesis, written by Margarita Teresa Almeida, and entitled Substructuring in Four Populations of African Descent, having been approved in respect to style and intellectual content, is referred to you for judgement.

We have read this thesis and recommend that it be approved.

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Date of Defense: July 24,1997
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I dedicate this thesis to my family and Clinton A. El-Ramey for all their support.

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ABSTRACT OF THE THESIS

SUBSTRUCTURING IN FOUR POPULATIONS OF AFRICAN DESCENT

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When a suspect's DNA profile is admitted into court as a match to evidence the probability of the perpetrator being another individual must be calculated from database allele frequencies. The two methods used for this calculation are phenotypic frequency and likelihood ratio. Neither of these calculations takes into account substructuring within populations. In these substructured populations the frequency of homozygotes increases and that of heterozygotes usually decreases. The departure from Hardy-Weinberg expectation in a sample population can be estimated using Sewall Wright's F_{ST} statistic. F_{ST} values were calculated in four populations of African descent by comparing allele frequencies at three short tandem repeat loci. This was done by amplifying the three loci in each sample using the Polymerase Chain Reaction and separating these fragments using polyacrylamide gel electrophoresis.

The gels were then silver stained and autoradiograms taken, from which allele frequencies were estimated. F_{ST} values averaged 0.007$\pm$ 0.005 within populations of African descent and 0.02$\pm$ 0.01 between white and black populations.

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I. INTRODUCTION

When a suspect's DNA profile is admitted into court as evidence the probability of the perpetrator being another individual must be calculated from database allele frequencies. There are two methods commonly used for this calculation. One method includes determining the phenotypic frequency. That is the probability of the phenotypic frequency is calculated from database allele frequencies for that population. The second method includes the determination of the likelihood ratio. Which is the ratio of the probability of a match if the DNA in the evidence sample and that from the suspect came from the same person to the probability of a match if it came from a different person. The same information is given in the calculations of phenotypic frequency and likelihood ratio because they are reciprocal(1-5).

Neither of these calculations takes into account substructuring of the populations. Since the suspect belongs to a subpopulation, it is possible to calculate phenotypic frequencies by assuming that substructuring is present. This would give a different calculation of phenotypic frequency or likelihood ratio(1,3-6).Inbreeding

is defined as the mating of two persons who are more closely related than if they were chosen at random.This can occur when marriage between close relatives takes place, such as, second cousin marriages. Alternatively it can occur because the entire population is divided into subpopulations showing positive assortive mating. Both forms of inbreeding cause an increase in the frequency of homozygotes and usually a decrease in the frequency of heterozygotes. The first type of inbreeding is denoted as F_{IS} or F_{IT}. They measure the decrease in heterozygosity of an individual within its subpopulation and the decrease in heterozygosity of an individual relative to the whole population, respectively. The second, the degree of relatedness in a subpopulation can be estimated by using Sewall Wright's F_{ST} statistic. F_{ST} measures the degree of excess homozygosity within a subpopulation in comparison with the total population(7,8). This population subdivision causes an increase in the frequency of homozygous genotypes and a decrease in the frequency of heterozygous genotypes(1,7,8).

F_{ST} values were calculated for all possible comparisons for 60 samples of each of the following populations: Broward

whites, Broward blacks, Bahamians, Jamaicans, and Trinidadians. The average F_{ST} value was found to equal 0.01 for these populations. This was done by determining the allele frequencies for three short tandem repeat loci for each sample in the populations used.

Short tandem repeats (STR's) are polymorphic loci consisting of 3-7 base pair repeats that occur in tandem in varying frequencies in eukaryotic organisms. The three loci used were HUMCSF1PO, HUMTPOX,and HUMTHO1. HUMCSF1PO (CSF1PO) is located on chromosome five at the site for colony stimulating factor-1 receptor gene and has a repeat sequence of 5'-AGAT-3' that occurs between 6-15 times, in most populations. HUMTPOX (TPOX) is located on chromosome two at the site of the human thyroid peroxidase gene and has a repeat sequence of 5'-AATG-3' that occurs between 6-13 times. HUMTHO1 (THO1) is located on chromosome eleven at the site for the human tyrosine hydroxylase gene and has a repeat sequence of 5'-AATG-3' that is repeated between 5-11 times. There is a common mutation in the THO1 STR that is not a true STR but since it so common it is used as an additional allele for identification purposes. This 9.3

allele repeat is, in fact, a 10 allele repeat with a base pair deletion(9,10,11).

These three STR loci were amplified using the polymerase chain reaction (PCR) separated by electrophoresis in a denaturing polyacrylamide gel. The alleles were then read from an autoradiogram of the silver stained gel.

II.MATERIALS AND METHODS

DNA EXTRACTION

1. Cut a .5 cm X .5 cm square of stain and place in a 1.5 ml microcentrifuge tube.
2. Add 400 µl stain extraction buffer and 10 µl proteinase K. Mix and spin in Eppendorf microcentrifuge 5415C for 1 minute at 10,000 rpm to force stain to bottom of the tube.
3. Incubate at 56 °C overnight.
4. Spin for 2 seconds to force condensation to bottom of tube.
5. Punch 3 holes in lid of tube in triangle formation, open tube and place cutting on lid and close and spin for 5 minutes at 10,000 rpm.
6. Remove lid and discard in biohazzardous container.

* Following two steps must be conducted in fume hood*

7. Add 500µl of phenol/chloroform/isoamyl alcohol, place new lid on tube, shake well by hand to achieve a milky emulsion in the tube, spin for 2 minutes at 10,000 rpm.

8. Transfer the aqueous phase (top layer) to another 1.5 ml microcentrifuge tube.

9. To the aqueous phase add 1.0 ml of cold absolute alcohol.

10. Mix by hand and place in -20 °C freezer for 30 minutes.

11. Spin tube for 20 minutes at 10,000 rpm.

12. Remove absolute alcohol by slow decantation.

13. Add 1 ml of room temperature 70% alcohol.

14. Spin for 5 minutes at 10,000 rpm.

15. Remove 70% alcohol by slow decantation being careful not to disturb pellet.

16. Place tube in Savant Speed-vac, to remove remaining alcohol, for approximately 20 minutes.

17. Resolubilize the DNA in 36 µl of TE overnight in 56 °C incubator.

or after step #7 continued with following protocol

8. Add aqueous layer to Microcon-100 (Amicon #42413) and bring up volume to top line with TE.

9. Spin in centrifuge at 5100 rpm for 10 minutes.

10. Dispense TE from reservoir and add more TE to filter to top line.

11. Spin in centrifuge at 5100 rpm for another 10 minutes.

12. Dispense entire reservoir, invert filter into sample reservoir and spin at 5100 rpm for 2 minutes.

13. Add approximately 20 µl of ultrapure water to DNA pellet.

14. Vortex briefly then centrifuge at 5100 rpm for 1 second. Can immediately go to yield gel.

YEILD GEL

1. Measure 1 gm of ultrapure agarose in 250 ml Erlenmeyer flask and 100 ml of TAE and microwave for 3 minutes or until agarose dissolves.

2. Add 10 µl of ethidium bromide and let gel mix cool to 56 °C.

3. Pour in 12 X 15 cm gel tray and place gel comb in place and let stand 30 minutes.

4. Place gel in gel tank and add TAE buffer so that gel is covered.

5. Add 6 µl of each of the following DNA standards to the first six wells: 500 ng , 250 ng, 125 ng, 63 ng, 31 ng, and 15 ng.

6. Add 4 µl of sample DNA and 2 µl of loading solution in the following wells.

7. Set voltage at 200 volts on power supply and run for approximately 20 minutes or till loading solution has run 1-2 cm from the origin.

8. Turn off power supply and remove gel from tank and view on Fotodyne ultraviolet light transluminator.

9. Photograph the gel with Polaroid #667 8.5 X 10.8 cm film. With red filter in place being careful not to expose eyes to UV light.

10. Using standards quantitate amount of DNA in sample.

11. Can go immediately to Amplification of sample or further quantitate using the Slot Blot procedure

SLOT BLOT

1. Determine number of samples to be blotted including the following DNA standards

(10 ng , 5 ng , 2.5 ng , 1.25 ng , .625 ng , .312 ng , .156 ng).

2. Add 150 µl of spotting solution for each sample and standard to microtiter plate.

3. Add 5 µl of each standard in its corresponding well.

4. Using yield gel quantitation add approximately 5 ng of sample DNA to its corresponding well.

5. Obtain 8 X 11 cm Biodyne B membrane and cut a notch on the bottom right corner for orientation and soak in wetting solution for 1 minute.

6. Place membrane on gasket of the Gibco BRL Convertible Filtration Manifold System and place top plate over membrane.

7. Turn on vacuum source, turn off sample vacuum and turn on clamp vacuum. Push down to insure tight seal.

8. Pipette 155 µl of sample and spotting solution mixture, as well as standards in the appropriate slots using a different pipette for each sample.

9. When all sample have been blotted turn on sample vacuum and leave on till all samples have been drawn through membrane and each slot has a uniform blue band.

10. Turn off clamp vacuum then vacuum source.

11. Disassemble slot blot apparatus and remove membrane.

12. Bake membrane between blotting paper in 56 °C incubator for at least one hour.

LABELING PROCEDURE FOR D17Z1

1. Place volume of probe in a 1.5 ml tube and bring volume up to 6µl with TNE then add water to final volume of 30 µl.

2. Heat tube at 95-100 °c for 2 minutes then immediately place in cooler for 5 minutes.

3. After 5 minutes, spin for 2 seconds to retrieve condensation and add the following in order:

10 µl 5X labeling Buffer

2 µl mixture of unlabeled dNTP's (ratio 1:1:1)

2 µl nuclease-free BSA

5 µl {C-^{32} P} dNTP (50uCi, 3000Ci/mmole)

5 units Klenow Enzyme

For a final volume of 50µl

4. Spin tube for 2 seconds and incubate for 2 hours.

5. Remove caps from sephadex columns and decant liquid.

6. Fill columns with TE and decant.

7. Place column on Lucite rack and fill with TE. Remove bottom plugs and allow TE to flow through the column into waste container.

8. Add entire labeled probe volume to the Sephadex column and allow to completely enter filter.

9. Add 400 µl TE to the Sephadex column and allow to run through into waste container.

10. Add 400 µl TE to column and collect the liquid as it is eluted. After probes are completed eluted dispose of columns.

11. Place 2 µl of probe into the 1.5 ml tube to determine total radioactivity.

12. Spin tube to be certain sample is located at bottom center where reading is taken with the Dupont Benchcount (model BC2000).

HYDRIDIZATION

1. Pre-hybridize the membrane with 20 ml of the pre-warmed hybridization solution in a crew cap hybridization tube with .5 ml herring sperm for 20-30 minutes in 65 °C rotisserie oven.

2. Combine 0.5 ml herring sperm DNA and the number of microliters calculated depending on the radioactivity reading from labeled probe.

3. Punch hole in cap to release pressure and place in 95 °C heat block for 5 minutes.

4. Immediately pipette the contents of the tube into 20 ml of 56 °C hybe solution, mix and add to the hybridization tube. Rotate overnight at 65 °C.

5. Pour off the hybridization solution and rinse membrane briefly in 2X SSC and discard solution.

6. Transfer membrane to a container and wash in 2X SSC: 0.5% SDS at 65 °C for 10 minutes.

7. Pour off solution and rinse with 2X SSC.

8. Lightly blot the membrane and wrap in Saran wrap.

9. In darkroom place a piece of Kodak Biomax MS Scientific Imaging Film in a film cassette.

10. Tape the covered membrane on top of film so that the DNA side is in contact with the film. Close the film cassette.

11. Expose the film overnight at -80°C.

12. Process the film in the Konica Medical Film Processor (QX-70).

CTT AMPLIFICATION USING PCR

1. Turn on Perkin Elmer Cetus Gene Amp 9600 PCR System approximately 30 minutes before use.

2. Determine volume of extracted sample needed from Slot Blot so that approximately 3 ng of DNA is added to reaction

mix, bring up to final volume of 5µl with nuclease free water so that a total volume of 5µl is added to each reaction tube.

3. Add 1µl of BSA (8 mg/ml) to DNA mixture.

4. To each microcentrifuge tube add 20 µl of master mix which contains the following:

14.85 µl of nuclease free water

2.5 µl 10X STR Buffer

2.5 µl 10X CTT Primer

0.15 µl Taq Polymerase

5. Last add the 5µl of DNA mixture to the corresponding test tube.

6. Always include a positive and negative control. The positive control contains 20µl of master mix and 5 µl of DNA mixture that is 2.5 µl of K562 DNA and 2.5 µl of nuclease free water. The negative control contains 20 µl of master mix and 5.0 µl of nuclease free water.

7. Mix each test tube using a vortex for approximately 2 seconds followed by centrifuging for approximately 2 seconds then place each test tube in the Perkin Elmer 9600 and amplify using the following protocol:

Pre-denature	96 °C	2 minutes	
Denature	94 °C	1 minute	
Anneal	64 °C	1 minute	for 10 cycles
Extend	70 °C	1.5 minutes	
Denature	90 °C	1 minute	
Anneal	64 °C	1 minute	for 20 cycles
Extend	70 °C	1.5 minutes	

PRODUCT GEL FOR VERIFICATION OF AMPLIFICATION

1. In an Erlenmeyer flask add 2 gm of ultra pure agarose and 100 ml of TBE and microwave for approximately 3 minutes or until agar is dissolved completely.

2. Add 10 µl of ethidium bromide to flask and let cool to 56 °C and pour into 12 X 15 cm gel tray, make sure well comb in place and let solidify (approximately 30 minutes).

3. Once solidified place in TBE electrophoresis tank and remove comb.

4. To the first well add 12 µl of $\phi\chi$174 as a marker to identify if triplex bands appear.

5. To each consecutive well add the following mixture: 10 µl of amplified product for each sample and 2 µl of loading solution without xylene cyanol.

6. Run gel at 100 volts for 60 minutes.

7. Once gel run is complete turn off power supply and view on Fotodyne UV light transilluminator.

8. Photograph the gel with Polaroid #667 8.5 X 10.8 cm film with red filter in place.

9. Using φχ174 as size standard, if three bands appear between base pair range of 118-310 then amplification was successful.

ANALYTICAL GEL

1. Prepare 4% or 6% (for the detection of the THO1 9.3 allele) denaturing polyacrylamide gel using the following:

Component	4%	6%
Urea	31.50g	31.50g
DH_2O	40.00ml	36.25ml
10X TBE	3.75ml	3.75ml
40% acrylamide:bis (19:1)	7.50ml	11.25ml

2. Filter gel solution through 0.2 micron filter (Nalgene tissue culture filter). Can be stored the light proof container at 4°for up to three months.

3. Treat the shorter gel plate with fresh binding solution (prepare just before treating plate) made with 1.5 ml of 0.5% acetic acid and 3 µl of bind silane.

4. Pour this mixture on cleaned shorter plate and spread with kim wipe in circular motion making sure to cover whole plate.

5. Let dry for 5 minutes and remove excess by wiping with 95% ethanol on kim wipe four times, if not gel will stick to both short and long plate.

6. Treat longer plate with Gel Slick. Add 3 ml of Gel Slick and spread with paper towel using circular motion.

7. Let dry for five minutes and remove excess with paper towel saturated with distilled water and let dry.

8. Assemble plates with treated sides facing each other separated by 0.4 mm side spacers and clamp with 4 clamps on each side leaving top and bottom unclamped.

9. Pour 32 ml of gel solution in squirt bottle and add 250 µl of ammonium persulfate and 25 µl TEMED and swirl to mix.

10. Immediately pour gel from top of plate being careful not to let bubbles form till solution reaches bottom of

plate and starts to drip out insert well comb at top on gel plates and clamp in place.

11. Let gel polymerize for at least one hour.

12. After gel has polymerized remove clamps and comb and shave away excess gel and clean plates with kim wipes saturated in deionized water.

13. Assemble plates in BRL model SA vertical electrophoresis apparatus , make sure top drain closed and add 0.5X TBE to top buffer chamber until well front is covered.

14. Make sure that buffer is not leaking between plates and apparatus and then add buffer to bottom chamber till it reach and slightly covers the bottom of the plates.

15. Using 100 cc syringe filled with buffer and remove air bubbles and top of gel and any small pieces of polyacrylamide.

16. Pre-run gel at 40 watts for approximately 40 minutes or until reaches 50 °C.

17. While gel is pre running prepare PCR samples by taken 2.5 µl of amplified product with 2.5 µl of STR 2X loading solution and do the same for STR ladder.

18. Denature sample and ladders for 2 minutes at 96 °C and immediately chill. Do this when gel pre- run is almost complete just before loading.

19. Once pre-run is complete at 3 µl of denatured sample mix in appropriate well, making sure that whole gel is loaded before it cools.

20. After gel is loaded with samples, ladders and positive and negative controls run at 40 watts for 55 minutes for 4%gel and 2 hours for 6% gel.

21. Once gel run in complete turn off power supply, drain top chamber and then remove gel plates.

22. Using spatula separate plates, gel should stick to the short plate.

23. Now gel is ready for staining so that STR alleles can be assigned to each sample using the ladder as guide.

24. A 6% Long Ranger Gel Solution was also used for the determination of the THO1 9.3 allele the following protocol was used.

25. The following mix was made prior to each gel run using Long Ranger Gel Solution:

10X TBE	3.2 ml
Urea	13.44 gm
Ultrapure water	qs 32 ml

50% Long Ranger 3.84 ml

26. Follow plate treatment as stated above.

27. Prior to pouring gel add 160 µl 10% ammonium persulfate and 16 µl Temed to gel mix.

28. Follow same directions as above to pour, pre-run and load gel.

29. Run at constant 40 watts for approximately 2 hours or till the second dye in the loading solution is about to run out of gel.

SILVER STAINING

1. Place gel in shallow plastic container in fix/stop solution for 20 minutes.

2. Pour fix/stop in another container for latter use.

3. Soak gel in deionized water for 2 minutes, repeat 2 more times discarding the water each time.

4. Soak gel in staining solution for 30 minutes then pour staining solution in silver precipitation container.

5. Rinse in deionized water again but only for 10 seconds.

6. Transfer gel to another shallow plastic container that is only used for developer solution and soak in developer for up to five minutes or until bands appear.

7. Pour previously used fix/stop into container and soak for 5 minutes along with developer.

8. Pour out developer and fix/stop and soak gel in deionized water for 2 minutes.

9. All of the staining step are conducted while container is rocking slowly.

10. Once all step are completely the plate with gel affixed is let dry overnight in hood.

EXPOSURE OF FILM

1. Once gel is dry, place on counter in darkroom with gel side up.

2. Place a piece of Kodak 20.3 X 25.4 Duplicating Film with emulsion side facing the gel.

3. Make sure that film makes full contact with gel by running hand over the film.

4. Turn gel over so that film in now under plate and expose film by turning on overhead fluorescent light for 2 seconds.

5. Now take film off of gel and process in Konica Medical Film Processor QX-70.

6. Once film is developed then the number of repeats for each sample for each loci can be determined by comparison with allelic ladder.

III.RESULTS

ALLELE ASSIGNMENT FOR BROWARD WHITES

SAMPLE#	CSF1PO	TPOX	THO1
BRW01	12,14	9,10	7,8
BRW02	12,15	8,11	9.3
BRW03	10,12	8,9	7,9.3
BRW04	10,12	11	7,9.3
BRW05	9,10	8,9	6
BRW06	11,12	8,10	7,8
BRW07	10,11	10,11	8,9.3
BRW08	10,11	8,10	8,9.3
BRW09	10	8,9	6,9.3
BRW10	11	8	6,7
BRW11	10,12	8	7
BRW12	11	8,10	9,9.3
BRW13	11,12	8,9	9.3
BRW14	10,12	8,11	7,10
BRW15	11,13	8	7,9
BRW16	11,12	8,10	7,8
BRW17	11,13	8,11	6,10
BRW18	11,12	11	7,9
BRW19	11,12	8,11	6,8
BRW20	11,12	8	7,8
BRW21	12,15	8	6,8
BRW22	11	8,9	6,7
BRW23	11,12	11,12	9.3
BRW24	10,12	11,12	7
BRW25	12	8	6,9
BRW26	9,10	8	8,9
BRW27	12,13	8,11	7,9.3
BRW28	11,12	8,11	7,9
BRW29	10,11	8	9,9.3
BRW30	11,12	8,9	7,9
BRW31	10,12	8,9	7,9.3

BRW32	11,12	8	6,9
BRW33	12	8,11	6,9
BRW34	11	8,11	9.3
BRW35	9,10	8,11	8,9
BRW36	11,12	8,11	6,9
BRW37	10,12	11	7,9
BRW38	11	8	6
BRW39	11,12	9,10	6,9.3
BRW40	11,12	8,11	7,9
BRW41	12	8,11	6,7
BRW42	11,12	9,11	7,9.3
BRW43	12,13	8,12	7,9.3
BRW44	10,11	9	6
BRW45	11	8,11	7,8
BRW46	11,12	8,12	8,9.3
BRW47	10,12	8,11	8,10
BRW48	11,12	9,11	6,9
BRW49	11,12	8,11	9
BRW50	9,11	8	6,8
BRW51	10,12	8,10	7,9
BRW52	12	8,9	6,9.3
BRW53	11,12	9,11	9.3
BRW54	11,12	10,11	7,9.3
BRW55	10	8,10	7,8
BRW56	9,11	8	6,9.3
BRW57	12	8,9	7,9.3
BRW58	11,12	8	7
BRW59	10	8,11	8,9.3
BRW60	10	10,11	8,9

ALLELE ASSIGNMENT FOR BROWARD BLACKS

SAMPLE#	CSF1PO	TPOX	THO1
BRB01	8,13	6,9	7
BRB02	10,11	11	6,7
BRB03	10,12	9,11	6,7
BRB04	10	8	7
BRB05	10	8,9	6,7
BRB06	8,14	7,8	6,7
BRB07	8,13	8,10	7,9
BRB08	10	8	6,7
BRB09	11	8,11	6,9.3
BRB10	10,12	11	7
BRB11	12	9,10	7
BRB12	10,12	8	7,8
BRB13	7,11	8,9	8,9.3
BRB14	10,12	8	7,8
BRB15	10,13	8	6,9
BRB16	10,11	6,11	8,9.3
BRB17	9,10	9,10	7,8
BRB18	9,12	9	7,8
BRB19	10,12	6,8	7,9
BRB20	11,12	8,10	7
BRB21	10,11	11	7
BRB22	10	9	6,8
BRB23	10,12	7,10	7
BRB24	10,11	7,11	8,9
BRB25	10,11	8,12	8,9
BRB26	10,12	8,9	7,9
BRB27	8,12	8,11	8,9
BRB28	10,11	7	6,7
BRB29	12	9,12	8,9
BRB30	10,12	9,10	7,8
BRB31	11,12	8,11	8,9
BRB32	12,14	9,10	7,8
BRB33	8,12	8,11	6,9
BRB34	10,12	6,8	7
BRB35	10,12	8,9	7
BRB36	10	9,10	6,8
BRB37	11	8,11	8
BRB38	9,12	8,11	8,9
BRB39	10,12	6,9	7,9
BRB40	9,12	8	7,9
BRB41	7,12	8,11	7,9

BRB42	9,10	6,10	7,8
BRB43	12,13	8,11	6,8
BRB44	12,13	11	7
BRB45	11,12	8,10	8,9
BRB46	9,11	10,11	7,8
BRB47	11,12	8	9
BRB48	10,11	8,9	6,7
BRB49	8,10	7,11	7,8
BRB50	10,12	8,11	6,8
BRB51	7,11	8	6,7
BRB52	10,11	11	6,9.3
BRB53	11	8,9	9.3
BRB54	10,13	10,11	7,9.3
BRB55	7,12	8,10	6
BRB56	11,12	12	7,9
BRB57	7,12	8	8
BRB58	10,13	7,9	8,9.3
BRB59	9	8,12	6,7
BRB60	10,12	8	7,9.3

ALLELE ASSIGNMENTS FOR BAHAMIANS

SAMPLE#	CSF1PO	TPOX	THO1
BAH01	10	8,9	6,9.3
BAH02	11,12	6,9	7,8
BAH03	10,12	9,11	8,9.3
BAH04	11,12	9,10	6,9
BAH05	13	9	8,9.3
BAH06	10,12	11	7
BAH07	8,12	10,11	7,8
BAH08	11,12	9	7,9.3
BAH09	10,11	6,11	6,8
BAH10	10	11	7,8
BAH11	8,13	8,12	7,8
BAH12	11,13	8,10	6,7
BAH13	10,12	11	7,8
BAH14	10,12	8,9	7,9
BAH15	12	9,11	7
BAH16	11,12	8	8,9
BAH17	10,11	9	7
BAH18	9,13	6,11	7,8
BAH19	11,12	9	6
BAH20	11,12	8,9	8,9
BAH21	7,10	10	7,8
BAH22	11,13	8,11	6,7
BAH23	11,12	7,8	7,9
BAH24	7,10	8,9	7,8
BAH25	12	9,10	7,8
BAH26	11	9	7,8
BAH27	11,12	9,12	8,9.3
BAH28	10	6,11	7
BAH29	7,15	6,9	8,9
BAH30	7,12	10,11	7
BAH31	10,12	8,9	7
BAH32	11,12	6,9	7,8
BAH33	8,10	9,11	7,9
BAH34	7,8	9,11	7,9
BAH35	11,12	8,9	8,10
BAH36	11	7,11	6,7
BAH37	10,11	8	8,9
BAH38	12	8,11	7
BAH39	11,12	10,11	9,10
BAH40	10,13	8,11	7,8
BAH41	7,12	8,11	7,9.3

BAH42	11	11	6,7
BAH43	11,12	6,11	6,8
BAH44	10,12	8,9	7,8
BAH45	11	6,11	9
BAH46	11,12	6,7	7,9
BAH47	7,10	6,8	7,9.3
BAH48	11,12	10,12	7,8
BAH49	10,12	7,10	7,9
BAH50	12	9	8,9
BAH51	12,13	12,13	6,7
BAH52	10,11	8,10	7,9
BAH53	10,11	9	8,9.3
BAH54	8,10	6,10	7,8
BAH55	12	8,11	7
BAH56	11	6,11	9.3
BAH57	9,11	8,11	6,7
BAH58	11,12	6,11	8,9
BAH59	11,12	6,11	6,7
BAH60	10,11	11	8

ALLELE ASSIGNMENT FOR JAMAICANS

SAMPLE#	CSF1PO	TPOX	THO1
JAM01	10,11	9,11	7,8
JAM02	10,11	8,9	6,7
JAM03	11,13	6,8	7,8
JAM04	12,13	9,11	7,9.3
JAM05	11	6	8
JAM06	12,13	9,10	7,9
JAM07	7,8	8,9	6,7
JAM08	10,12	8,9	6,9
JAM09	11,12	9	7,8
JAM10	10	9,11	7,8
JAM11	10	8	9,10
JAM12	12	8	7
JAM13	10	8,10	7
JAM14	10,12	8,11	6,8
JAM15	8,11	9,11	7
JAM16	11,13	8,12	7,9.3
JAM17	12,14	8,9	7,8
JAM18	10,11	8	7,9
JAM19	9,10	10,11	6,9
JAM20	7,10	8,11	7
JAM21	8,9	9,10	7,8
JAM22	8,10	8,9	6,9
JAM23	7,11	6,9	6,9
JAM24	9,11	9	6,8
JAM25	11,12	8,11	7,8
JAM26	10,12	6,8	7,8
JAM27	11,12	8	8,10
JAM28	10,12	8,9	8,9
JAM29	12	9,10	7,8
JAM30	10,13	8,9	6,7
JAM31	10,11	8,11	6,7
JAM32	11,12	11	6,8
JAM33	9,10	8,11	8
JAM34	11,13	8	8,9.3
JAM35	7,13	6,8	6,7
JAM36	8,12	11	6,7
JAM37	10	10,11	6,7
JAM38	9,10	8,10	6,9
JAM39	8,12	8	7,9
JAM40	10	10,11	7
JAM41	11,12	6,9	7

JAM42	11,12	8,9	8
JAM43	11,12	9	6,7
JAM44	7,12	9	6,9.3
JAM45	7,12	8	7,9
JAM46	7,10	11,12	7
JAM47	11,12	9,10	7,8
JAM48	9,10	9	7
JAM49	7,12	8,11	7
JAM50	11,12	9,11	7,8
JAM51	11,12	11,12	6,7
JAM52	7,8	6,9	7,9
JAM53	8	8,11	6,8
JAM54	10,13	9,11	6,7
JAM55	9,11	8,9	7
JAM56	10,11	6,11	6,8
JAM57	8,11	8,9	8
JAM58	7,11	9	7
JAM59	9,13	9,10	7,9
JAM60	7,10	7,12	7,9

ALLELE ASSIGNMENTS FOR TRINIDADIANS

SAMPLE#	CSF1PO	TPOX	THO1
TRI01	12	9,12	7,8
TRI02	8,12	11,12	7,9
TRI03	11,14	9,12	7
TRI04	10,11	10,11	7,9
TRI05	11	6,8	7,8
TRI06	10,11	8,11	6,7
TRI07	10	8,11	6,9.3
TRI08	12	10,11	7,9
TRI09	12	8,9	7
TRI10	8,12	11	6,9
TRI11	10,12	8,10	6,7
TRI12	10,11	8,9	9,9.3
TRI13	8,13	7,8	6,9
TRI14	11,12	12,13	6,8
TRI15	8,11	8,11	6,8
TRI16	13	9	7,8
TRI17	13	8,10	8
TRI18	10	11	6,7
TRI19	7,10	6,8	9,10
TRI20	10,11	8,11	6,7
TRI21	8	8,10	6,7
TRI22	11	8,9	7,8
TRI23	10,11	8	7,8
TRI24	10,12	9,12	7,9
TRI25	7,12	8,11	7,9
TRI26	8,12	6,8	8,10
TRI27	8,9	7,10	7
TRI28	10,13	8	6,8
TRI29	10	6,11	7,8
TRI30	7,10	8	7
TRI31	7,10	6,11	7,9
TRI32	10,12	9,11	6,9.3
TRI33	8,12	6,8	7
TRI34	12	8,11	9
TRI35	8,12	6,8	7,9
TRI36	10,12	10,11	6,8
TRI37	10,12	10,11	7,8
TRI38	7,11	8,11	7,8
TRI39	10	8,10	6,8
TRI40	10,11	9	7,8
TRI41	8,10	8	7,9

TRI42	9,12	9,11	6,8
TRI43	8,10	8,10	6,7
TRI44	8,12	8,9	7,9
TRI45	10,11	6,9	7
TRI46	7,12	11,12	8,9
TRI47	10,11	10,11	8,9
TRI48	11	9,11	7,8
TRI49	11,12	8,11	8,9
TRI50	10,12	11	7,8
TRI51	9,10	8,9	6
TRI52	10,12	8	7
TRI53	8,12	8	7,8
TRI54	11	9,10	8,9.3
TRI55	12,13	8,11	6
TRI56	12	8,11	6
TRI57	10	8,11	8,9
TRI58	12	9,11	6,9
TRI59	11,12	8,11	6,9.3
TRI60	11,12	8,11	8

F_{ST} Values for CSF

	BB	BAH	JAM	TRI
BW	0.0154	0.0041	0.0179	0.0160
BB		0.0084	0.0040	0.0026
BAH			0.0065	0.0064
JAM				0.0033

F_{ST} Values for TPOX

	BB	BAH	JAM	TRI
BW	0.0085	0.0410	0.0240	0.0097
BB		0.0158	0.0081	0.0021
BAH			0.0081	0.0125
JAM				0.0097

F_{ST} Values for THO1

	BB	BAH	JAM	TRI
BW	0.0163	0.0203	0.0255	0.0166
BB		0.0017	0.0018	0.0198
BAH			0.0038	0.0055
JAM				0.0039

Average F_{ST} Values for Loci

CSF	.0085
TPOX	.0140
THO1	.0115

Average F_{ST} Values by Population per Loci

Population	CSF1PO	TPOX	THO1
BW	.0134 sd.0063	.0208 sd.0152	.0197 sd.0043
ADP*	.0052 sd.0022	.0094 sd.0046	.0061 sd.0069

*ADP= African descent populations
sd.= standard deviation

IV.DISCUSSION

This research was conducted to determine the F_{ST} values for four populations of African descent in a PCR based system of indentification. These F_{ST} estimates may be significant because forensic DNA match calculations of phenotypic frequency and likelihood ratios do not take into account population substructuring (1-5). F_{ST} values greatly affect probability calculations when alleles are rare, but alter estimates to a lesser extent when alleles are common. For example, when there are is an allele presnt with a frequency of 0.01 in a given population the probability (p^2) of it occurring as a homozygote is 0.0001 or 1 out of 10,000. If an F_{ST} value of .01 is used in the calculation $p^2 + p(1-p)F_{ST}$ the match probability value changes to 0.0002 or 1 out of 5,000. Using an F_{ST} value of .03, which is recommended by the National Research Council for a conservative estimate of F_{ST} in a PCR based system, the match probability changes to 0.0004 or 1 out of 2,500(1). For a common allele with a frequency of 0.3 the probability (p^2) is .0.09 or 1 out of 11.11. Using F_{ST} values of 0.01 and

0.03 the probability changes to 0.0921 or 1 out of 10.86 and 0.0963 or 1 out of 10.38 , respectively. Average F_{ST} estimates for all populations at STR locus CSF1PO equals 0.0085, for TPOX it is 0.0140 and for THO1 it is 0.0115. The average F_{ST} value for Broward whites compared to African descent populations at CSF1PO equals .0134, at TPOX it equals 0.0208 and at THO1 it equals 0.0061. This is in agreement with the general genetic observation that there is more substructuring between races than within races.

Since replicate allele frequencies effect F_{ST} values the samples used were carefully chosen. They were selected from Broward Sheriff's Office database of Broward whites, Broward blacks, Bahamians, Jamaicans, and Trinidadians. Sixty samples from each population were chosen based on the number of restriction fragment length polymorphisms (RFLP) sized in a previous study(12). All samples were sized at a minimum of four RFLP's. This ensured that no there were no samples repeated.

To test the significance of the F_{ST} differences observed additional statistical tests were performed. The G -test results (Appendix XI) indicated that Broward whites and

African descent populations are not homogeneous enough to be considered the same but all the African descent populations are homogeneous enough to be considered the same population(13). χ^2 values (Appendix XII) suggest that genotype can be predicted from allele frequencies in Broward whites compared to African descent populations but not between African descent populations. A T -test was conducted in order to determine whether the average F_{ST} values were significant different between Broward whites and African descent populations. T values (Appendix XII & XIV) suggest that the F_{ST} average between Broward whites and African descent populations are significantly different and therefore the populations are significantly different yet the average for African descent populations are not significantly diferent and therefore the populations are not significantly different.

The National Research Council suggested that for PCR based systems a conservative F_{ST} value of .03 may be used. For VNTR systems a value of .01 is a sufficient correction for subdivision. This is true for the populations used in this thesis(12,15). This study determined that the use of F_{ST}

equal to 0.01 is sufficient for a PCR based system in the African descent populations used.

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APPENDIX I

REAGENTS USED

0.5% Acetic Acid

Glacial acetic acid	50 ml
dH_2O	950 ml

0.5% Acetic Acid in 95% Ethanol

Glacial acetic acid	1 ml
95% Ethanol	199 ml

40% Acrylamide:bis

Acrylamide	380 g
Bisacrlamide	20 g

Dissolve above in 500ml d H_2O

then bring up to volume of 1L

Ammonium Persulfate

ammonium persulfate	0.5 g
dH_2O	5 ml

Store in freezer in 250 µl

aliquots

BSA

Bovine serum albumin	8 mg
Nuclease free water	1 ml

CTT Primer

Obtained from Promega in the

Geneprint STR Triplex CTT kit

Developing Solution

37% Formaldehyde	1.5 ml
Sodium Thiosulfate	200 µl
dH_2O	1000 ml
Sodium Carbonate	30 g

70% Ethanol

Reagent alcohol	700 ml
dH_2O	300 ml

Ethidium Bromide

dH_2O	40 ml
Ethidium Bromide	0.2 g

Fix/Stop

Glacial acetic acid	100 ml
DH_2O	900 ml

Herring Sperm DNA

Herring Sperm DNA	500mg

q.s. to 50ml of dH_2O

Denature in boiling water
for 5 minutes, aliqout 500 µl
in screw cap tubes

Hybridization Solution

20X SSPE	250 ml
20% SDS	25 ml
dH_2O	725 ml

Warm solution before use to ensure solids dissolved

Loading Buffer (Yield Gel)

Tris 0.5M	1.0 ml
TE	5.0 ml
Ficoll	1.0 g
dH_2O	4.0 ml
Bromophenol Blue	0.025 g

Phenol/Chloroform/Isoamyl Alcohol

Phenol	340 ml
Chloroform	340 ml
Isoamyl alcohol	13.6 ml
8-Hydroxyquinoline	0.68 g

Store in brown bottles

Prewetting Solution

Sodium Hydroxide	16 g
EDTA	9.3 g

Dissolve sodium hydroxide in 1 L

Of water to yield 0.4N Sodium Hydroxide
Dissolve EDTA in 1 L of 0.4N Sodium Hydroxide

Proteinase K

Proteinase K	500 mg
dH_2O	25 ml

Aliqout in 250 µl and freeze

SDS

SDS	200 g
dH_2O	700 ml

Heat to 65 °C to dissolve, adjust volume to 1 L

Sodium Thiosulfate

Sodium Thiosulfate	5 g
dH_2O	500 ml

Spotting Solution

0.4N Sodium Hydroxide/ 25mM EDTA	100 ml

Sprinkle with bromophenol blue

SSC

Sodium Chloride	526 g
Na_3 Citrate:2 H_2O	265 g
dH_2O	2400 ml

pH to 7.0 with HCl, q.s to 3 L

SSPE

Sodium Chloride	841.6 g
Sodium Phoshate	96 g

pH to 7.0 with 10M Sodium Hydroxide

0.5M EDTA	160 ml
dH_2O	4 L

Stain Extraction Buffer

10 mM Tris	1.21 g
0.1 M NaCl	5.84 g
0.01M $Na_2EDTA\text{-}2H_2O$	3.72 g

Dissolve Tris and NaCl in 500 ml dH_2O, adjust pH to 8.0 with NaOH. Add $Na_2EDTA\text{-}2H_2O$, add 100 ml 20% SDS. Adjust volume to 1 L with dH_2O.

Staining Solution

Silver Nitrate	1 g
dH_2O	100 ml
37% Formaldehyde	1.5 ml

STR Buffer

Obtained from Promega in

Geneprint STR Triplex CTT kit

STR Loading Solution

Obtained from Promega in

Geneprint STR Triplex CTT kit

50% TAE

Tris base	242 g
Glacial acetic acid	57.1 ml
0.5M EDTA	100 ml

Add Tris base and EDTA to 500 ml Of dH_2O. Add glacial acetic acid, q.s to 1000ml with dH_2O.

10X TBE

Tris base	107.8 g
EDTA	7.44 g
Boric acid (approx.)	55.0 g

Dissolve Tris base and EDTA in 800 ml dH_2O. Slowly add boric acid and monitor pH until obtain pH of 8.3, q.s to 1 L with H_2O.

TE

Tris base	1.21 g
EDTA	0.037 g

Dissolve Tris base and EDTA in 900 ml dH_2O. Adjust to pH 7.5 with HCl, q.s to 1 L with dH_2O.

TNE

Tris base	0.121 g
NaCl	0.584 g
$Na_2EDTA\text{-}2H_2O$	0.037 g

Dissolve in 80 ml dH_2O, adjust to pH 8.0 With NaOH, q.s to 1 L with dH_2O.

φχ174

$\phi\chi$174 RF (HAE Fragment) to a concentration of 100ng/6µl.

APPENDIX II

YIELD GEL RESULTS FOR BROWARD WHITES

SAMPLE #	DNA (ng)	SAMPLE #	DNA (ng)
BRW01	15<	BRW31	15<
BRW02	0	BRW32	15
BRW03	250	BRW33	15
BRW04	15	BRW34	15
BRW05	0	BRW35	31
BRW06	0	BRW36	63
BRW07	15	BRW37	0
BRW08	15<	BRW38	15
BRW09	63	BRW39	0
BRW10	31	BRW40	15
BRW11	0	BRW41	15<
BRW12	15<	BRW42	15
BRW13	250	BRW43	15
BRW14	15	BRW44	15
BRW15	0	BRW45	31
BRW16	31	BRW46	15
BRW17	31	BRW47	15
BRW18	15	BRW48	15<
BRW19	15	BRW49	15<
BRW20	63	BRW50	0
BRW21	30	BRW51	15
BRW22	15<	BRW52	31
BRW23	15<	BRW53	15
BRW24	15	BRW54	250
BRW25	15	BRW55	31
BRW26	15<	BRW56	0
BRW27	15<	BRW57	0
BRW28	15<	BRW58	15
BRW29	15<	BRW59	31
BRW30	0	BRW60	15

YIELD GEL RESULTS FOR BROWARD BLACKS

SAMPLE #	DNA (ng)	SAMPLE #	DNA (ng)
BRB01	500	BRB31	125
BRB01	15<	BRB32	250
BRB03	0	BRB33	125
BRB04	0	BRB34	63
BRB05	500	BRB35	125
BRB06	125	BRB36	0
BRB07	63	BRB37	250
BRB08	15	BRB38	15
BRB09	15	BRB39	125
BRB10	15<	BRB40	250
BRB11	15<	BRB41	125
BRB12	15	BRB42	250
BRB13	15	BRB43	15
BRB14	63	BRB44	31
BRB15	15	BRB45	125
BRB16	0	BRB46	7.5
BRB17	0	BRB47	63
BRB18	15	BRB48	15
BRB19	125	BRB49	15
BRB20	15	BRB50	63
BRB21	15<	BRB51	125
BRB22	0	BRB52	0
BRB23	31	BRB53	63
BRB24	15	BRB54	31
BRB25	0	BRB55	500>
BRB26	125	BRB56	15
BRB27	31	BRB57	500
BRB28	63	BRB58	15<
BRB29	15<	BRB59	31
BRB30	125	BRB60	15<

YIELD GEL RESULTS FOR BAHAMIANS

SAMPLE #	DNA (ng)	SAMPLE#	DNA (ng)
BAH01	7.5	BAH31	63
BAH02	31	BAH32	15
BAH03	15<	BAH33	63
BAH04	7.5	BAH34	15
BAH05	0	BAH35	15<
BAH06	15<	BAH36	15
BAH07	15<	BAH37	15<
BAH08	15<	BAH38	7.5
BAH09	0	BAH39	15<
BAH10	15	BAH40	0
BAH11	31	BAH41	15
BAH12	31	BAH42	15
BAH13	31	BAH43	15
BAH14	15<	BAH44	15
BAH15	31	BAH45	15<
BAH16	31	BAH46	15
BAH17	31	BAH47	15
BAH18	15	BAH48	15
BAH19	15	BAH49	31
BAH20	15	BAH50	15
BAH21	15<	BAH51	15<
BAH22	0	BAH52	15<
BAH23	0	BAH53	15
BAH24	0	BAH54	15
BAH25	15<	BAH55	15<
BAH26	15<	BAH56	0
BAH27	15<	BAH57	15<
BAH28	15	BAH58	15
BAH29	0	BAH59	15
BAH30	0	BAH60	22.5

YIELD GEL RESULTS FOR JAMAICANS

SAMPLE #	DNA (ng)	SAMPLE #	DNA (ng)
JAM01	31	JAM31	63
JAM02	31	JAM32	15
JAM03	31	JAM33	0
JAM04	63	JAM34	15<
JAM05	15	JAM35	15
JAM06	15<	JAM36	15<
JAM07	7.5	JAM37	15
JAM08	15	JAM38	15
JAM09	15	JAM39	31
JAM10	15	JAM40	250
JAM11	15<	JAM41	15
JAM12	15	JAM42	31
JAM13	15	JAM43	125
JAM14	15<	JAM44	125
JAM15	31	JAM45	31
JAM16	0	JAM46	63
JAM17	7.5	JAM47	63
JAM18	15	JAM48	125
JAM19	15	JAM49	125
JAM20	31	JAM50	125
JAM21	63	JAM51	250
JAM22	125	JAM52	125
JAM23	15	JAM53	125
JAM24	15	JAM54	31
JAM25	15	JAM55	31
JAM26	15	JAM56	15<
JAM27	15<	JAM57	15
JAM28	63	JAM58	63
JAM29	125	JAM59	15
JAM30	31	JAM60	63

YIELD GEL RESULTS FOR TRINIDADIANS

SAMPLE #	DNA (ng)	SAMPLE #	DNA (ng)
TRI01	15	TRI31	0
TRI02	0	TRI32	63
TRI03	31	TRI33	125
TRI04	15	TRI34	31
TRI05	15	TRI35	15<
TRI06	0	TRI36	15
TRI07	31	TRI37	31
TRI08	0	TRI38	0
TRI09	7.5	TRI39	0
TRI10	0	TRI40	15<
TRI11	15<	TRI41	63
TRI12	15<	TRI42	31
TRI13	31	TRI43	63
TRI14	7.5	TRI44	63
TRI15	15<	TRI45	15<
TRI16	15	TRI46	7.5
TRI17	0	TRI47	15
TRI18	0	TRI48	15<
TRI19	7.5	TRI49	15<
TRI20	15<	TRI50	15
TRI21	0	TRI51	15
TRI22	0	TRI52	125
TRI23	0	TRI53	15<
TRI24	15<	TRI54	15<
TRI25	0	TRI55	31
TRI26	0	TRI56	15<
TRI27	15	TRI57	15<
TRI28	15	TRI58	250
TRI29	15	TRI59	15
TRI30	0	TRI60	63

APPENDIX III

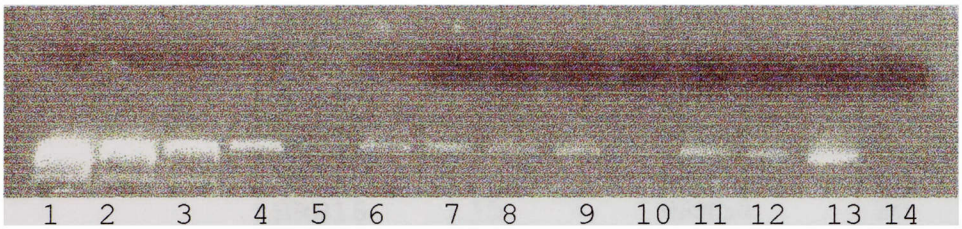

FIGURE 1: Scan of polaroid of yield gel. Lanes 1-6 contain the following standards in ng of DNA: 500, 250, 125, 63, 15, 30. Lanes 8-13 contain samples

APPENDIX IV

SLOT BLOT RESULTS FOR BROWARD WHITE

SAMPLE #	DNA (ng)	SAMPLE #	DNA (ng)
BRW02	10	BRW25	10
BRW03	30	BRW26	15
BRW04	30	BRW30	10
BRW05	10	BRW32	20
BRW06	30	BRW35	2.5
BRW09	30	BRW40	15
BRW11	5	BRW41	5
BRW15	1.25	BRW51	25
BRW16	15	BRW58	20
BRW18	10	BRW59	30
BRW19	10	BRW60	25

SLOT BLOT RESULTS FOR BROWARD BLACKS

SAMPLE #	DNA (ng)	SAMPLE #	DNA (ng)
BRB02	10	BRB27	20
BRB03	7.5	BRB28	30
BRB04	5	BRB34	10
BRB06	10	BRB35	40
BRB08	10	BRB36	5
BRB10	10	BRB37	60
BRB11	15	BRB38	10
BRB12	10	BRB39	10
BRB14	30	BRB40	30
BRB15	10	BRB41	30
BRB16	30	BRB43	10
BRB17	30	BRB45	30
BRB18	20	BRB47	20
BRB20	10	BRB50	20
BRB22	15	BRB51	30
BRB23	15	BRB55	20
BRB24	5	BRB58	15
BRB25	20	BRB60	10

SLOT BLOT RESULTS FOR BAHAMIANS

SAMPLE #	DNA (ng)	SAMPLE #	DNA (ng)
BAH02	5	BAH28	10
BAH06	.625	BAH29	30
BAH08	2.5	BAH30	30
BAH09	2.5	BAH31	10
BAH10	2.5	BAH32	5
BAH11	5	BAH33	2.5
BAH12	10	BAH34	10
BAH13	5	BAH36	10
BAH15	5	BAH41	20
BAH16	5	BAH42	20
BAH17	5	BAH43	20
BAH18	10	BAH44	20
BAH19	5	BAH46	20
BAH20	5	BAH47	20
BAH22	10	BAH48	20
BAH23	5	BAH49	20
BAH24	2.5	BAH50	20
BAH25	15	BAH53	10
BAH27	15	BAH54	5

SLOT BLOT RESULTS FOR JAMAICANS

SAMPLE #	DNA (ng)	SAMPLE#	DNA (ng)
JAM01	10	JAM35	30
JAM02	10	JAM37	30
JAM04	10	JAM38	5
JAM05	10	JAM39	30
JAM08	15	JAM40	20
JAM09	5	JAM43	20
JAM11	5	JAM44	20
JAM13	5	JAM45	63
JAM14	5	JAM46	20
JAM15	10	JAM47	20
JAM18	15	JAM48	20
JAM19	5	JAM49	20
JAM27	15	JAM50	20

JAM28	20	JAM51	20
JAM30	15	JAM52	20
JAM32	20	JAM53	20
JAM34	30		

SLOT BLOT RESULTS FOR TRINIDADIANS

SAMPLE #	DNA (ng)	SAMPLE #	DNA (ng)
TRI01	10	TRI21	15
TRI03	10	TRI25	15
TRI04	10	TRI26	2.5
TRI07	10	TRI27	10
TRI10	5	TRI28	5
TRI12	10	TRI29	10
TRI13	10	TRI43	20
TRI17	10	TRI44	30

APPENDIX V

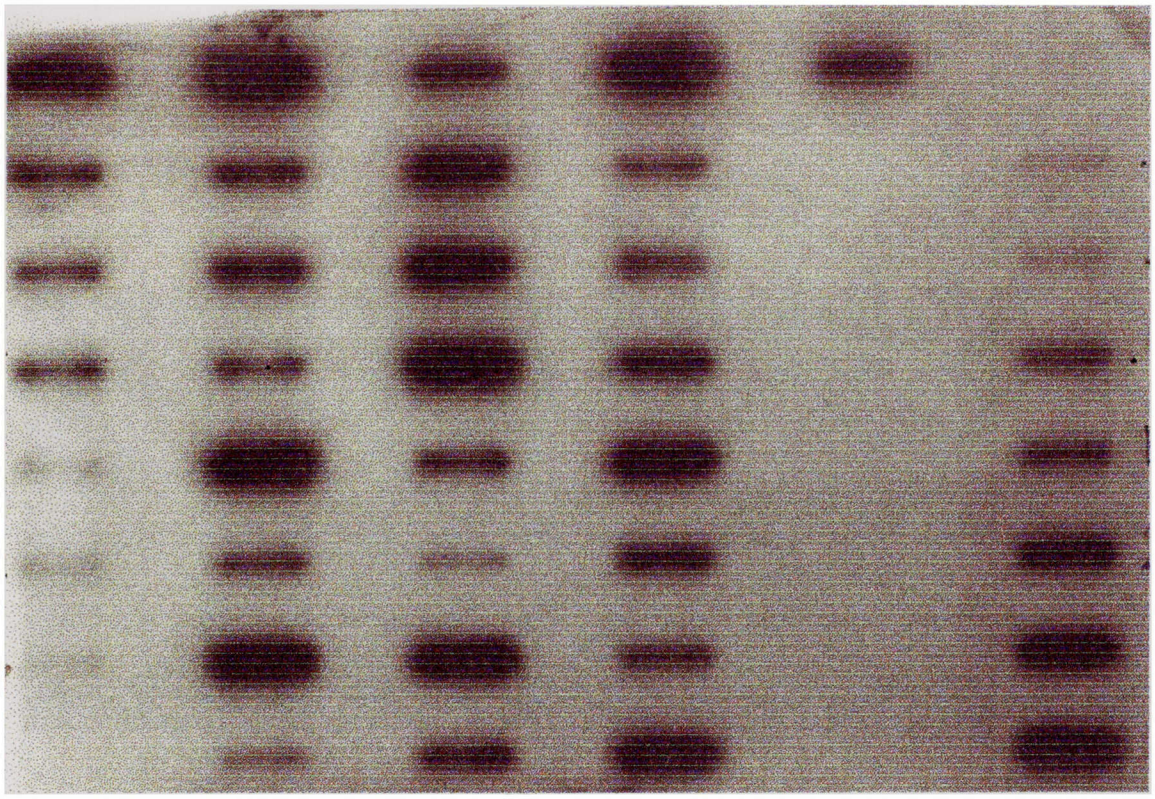

FIGURE 2:Scan of autoradiogram of Slot Blot for DNA sample quantitation. Lanes 1 and 6 standards in ng of DNA:10, 5,2.5,1.25,.625,.313,.156 in opposite order. Lanes 2-5 contain samples.

APPENDIX VI

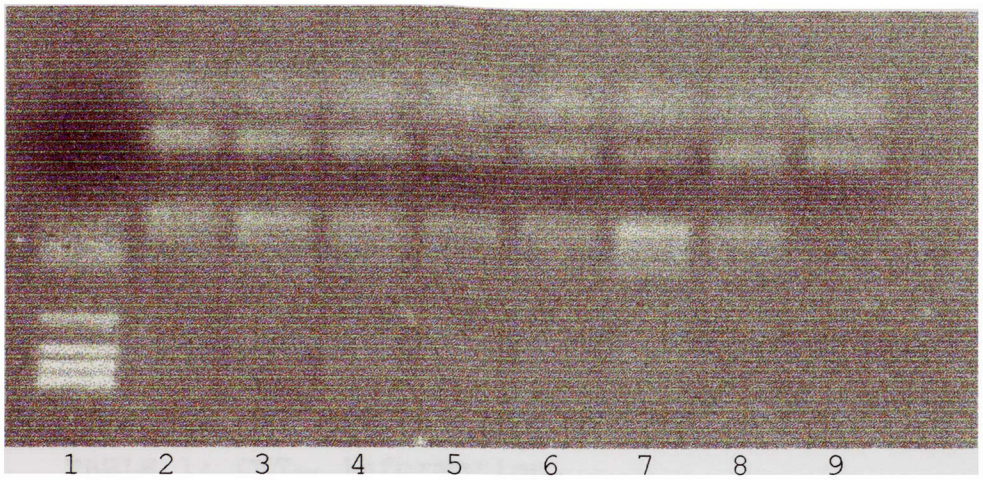

FIGURE 3: Scan polaroid of product gel. Lane 1 contains size marker φχ174. Lanes 2-8 contain samples. Lanes 9 contains negative control. All samples amplified.

APPENDIX VII

STR	CORE REPEAT	# OF CORE REPEATS	CHROMO-SOME SITE	CHROMO-SOME LOCATION	ALLELIC LADDER RANGE (bp)
CSF1PO	AGAT	7-15	CSF-1 RECEPTOR GENE	5	299-323
TPOX	AATG	6-13	THYROID PEROXID-ASE GENE	2	232-248
THO1	AATG	5-11	TYROSINE HYDROXY-LASE GENE	11	179-203

TABLE 1: CTT_{TM} Information.

APPENDIX VIII

ALLELE FREQUENCIES FOR CSF

CSF	BW	BB	BAH	JAM	TRI
15	2	0	1	0	0
14	1	2	0	1	1
13	4	7	8	9	7
12	44	33	37	26	34
11	40	22	35	26	23
10	24	37	25	29	32
9	5	8	2	8	3
8	0	6	5	10	14
7	0	5	7	11	6

ALLELE FREQUENCIES FOR TPOX

TPOX	BW	BB	BAH	JAM	TRI
13	0	0	1	0	1
12	4	5	4	4	6
11	30	25	32	23	32
10	11	13	12	10	12
9	16	20	31	36	18
8	59	44	22	37	41
7	0	7	4	1	2
6	0	6	14	9	8

ALLELE FREQUENCIES FOR THO1

THO1	BW	BB	BAH	JAM	TRI
11	0	0	0	0	0
10	3	0	2	2	2
9.3	28	9	10	4	5
9	19	18	17	14	20
8	17	27	31	27	28
7	31	47	48	52	41
6	22	19	12	21	24
5	0	0	0	0	0

APPENDIX IX

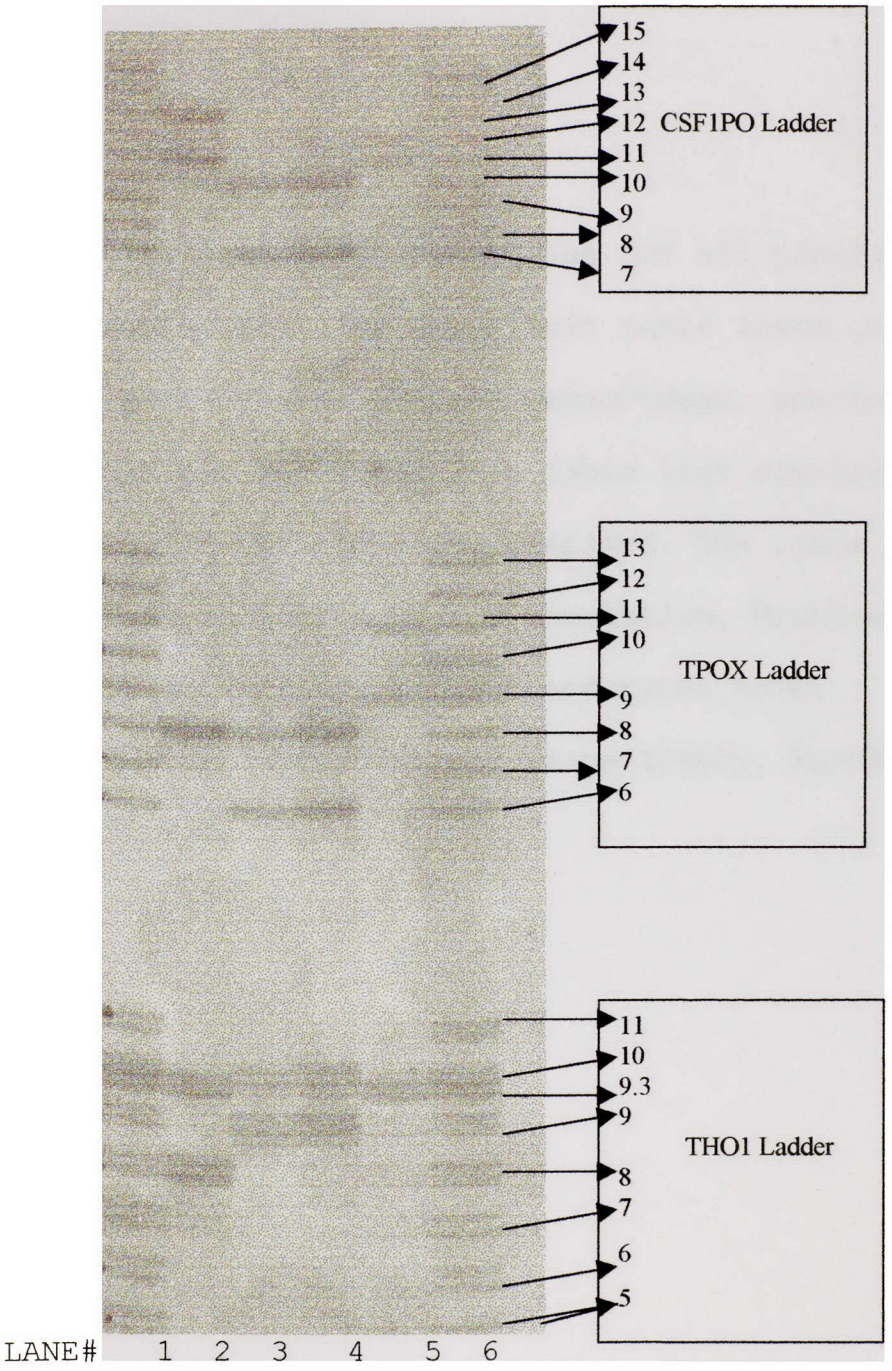

FIGURE 4 :Scan of 6% Long Ranger gel autoradiogram. Lane # 1 and 6 contain ladders. Lanes 2-5 contain samples.

APPENDIX X

Pages 59-88 : Effect of Substructuring for all populations for all three STR loci. The upper left table lists alleles, frequencies, percentages, squared percentages, and totals for the population. The upper left table list similar data for the population that is being compared. The lower left table is the theoretical combined population. H_T and H_S are heterozygosity estimates for the theoretical total population and the subpopulations respectively. $F_{ST}=(H_T-H_S)/H_T$.

CSF n=120	BW	F of BW	F2 BW	BB	F of BB	F2 BB
15	2	0.016666	2.77778E-04	0	0	0
14	1	0.008333	6.94444E-05	2	0.016666667	2.77778E-04
13	4	0.033333	0.001111111	7	0.058333333	0.003402778
12	44	0.366666	0.134444444	33	0.275	0.075625
11	40	0.333333	0.111111111	22	0.183333333	0.033611111
10	24	0.2	0.04	37	0.308333333	0.095069444
9	5	0.041666	0.001736111	8	0.066666667	0.004444444
8	0	0	0	6	0.05	0.0025
7	0	0	0	5	0.041666667	0.001736111
	120	1	0.28875	120	1	0.216666667

CSF H_s Values	BW	BB
	0.71125	0.783333

CSF	F BW+BB	F/240	F2(BW+BB)
15	2	0.008333	1
14	3	0.0125	0.00015625
13	11	0.045833	0.002100694
12	77	0.320833	0.102934028
11	62	0.258333	0.066736111
10	61	0.254166	0.064600694
9	13	0.054166	0.002934028
8	6	0.025	0.000625
7	5	0.020833	4.34028E-04
	240	1	1.240520833

Ht=	0.759
Hs=	0.747291
Fst=	0.015425

CSF	BW	F BW	F2 BW
15	2	0.016666	2.77778E-04
14	1	0.008333	6.94444E-05
13	4	0.033333	0.001111111
12	44	0.366666	0.134444444
11	40	0.333333	0.111111111
10	24	0.2	0.04
9	5	0.041666	0.001736111
8	0	0	0
7	0	0	0
	120	1	0.28875

BAH	F BAH	F2 BAH
1	0.008333333	6.94444E-05
0	0	0
8	0.066666667	0.004444444
37	0.308333333	0.095069444
35	0.291666667	0.085069444
25	0.208333333	0.043402778
2	0.016666667	2.77778E-04
5	0.041666667	0.001736111
7	0.058333333	0.003402778
120	1	0.233472222

CSF Hs values	BW	BAH
	0.71125	0.766527

CSF	BW+BAH	F BW+BAH	F2 BW+BA
15	3	0.0125	0.00015625
14	1	0.004166	1.73611E-05
13	12	0.05	0.0025
12	81	0.3375	0.11390625
11	75	0.3125	0.09765625
10	49	0.204166	0.041684028
9	7	0.029166	8.50694E-04
8	5	0.020833	4.34028E-04
7	7	0.029166	8.50694E-04
	240	1	0.258055556

Ht= 0.741944
Hs= 0.738888
Fst= 0.004118

CSF	BW	F BW	F2 BW
15	2	0.016666	2.77778E-04
14	1	0.008333	6.94444E-05
13	4	0.033333	0.001111111
12	44	0.366666	0.134444444
11	40	0.333333	0.111111111
10	24	0.2	0.04
9	5	0.041666	0.001736111
8	0	0	0
7	0	0	0
	120	1	0.28875

JAM	F JAM	F2 JAM
0	0	0
1	0.008333333	6.94444E-05
9	0.075	0.005625
26	0.216666667	0.046944444
26	0.216666667	0.046944444
29	0.241666667	0.058402778
8	0.066666667	0.004444444
10	0.083333333	0.006944444
11	0.091666667	0.008402778
120	1	0.177777778

CSF	BW	JAM
Hs	0.71125	0.822222
values		

CSF	F BW+JA	F/240	F2
15	2	0.008333	6.94444E-05
14	2	0.008333	6.94444E-05
13	13	0.054166	0.002934028
12	70	0.291666	0.085069444
11	66	0.275	0.075625
10	53	0.220833	0.048767361
9	13	0.054166	0.002934028
8	10	0.041666	0.001736111
7	11	0.045833	0.002100694
	240	1	0.219305556

Ht= 0.780694
Hs= 0.766736
Fst= 0.017879

CSF	BW	F BW	F2 BW
15	2	0.016666	2.77778E-04
14	1	0.008333	6.94444E-05
13	4	0.033333	0.001111111
12	44	0.366666	0.134444444
11	40	0.333333	0.111111111
10	24	0.2	0.04
9	5	0.041666	0.001736111
8	0	0	0
7	0	0	0
	120	1	0.28875

TRI	F TRI	F2 TRI
0	0	0
1	0.008333333	6.94444E-05
7	0.058333333	0.003402778
34	0.283333333	0.080277778
23	0.191666667	0.036736111
32	0.266666667	0.071111111
3	0.025	0.000625
14	0.116666667	0.013611111
6	0.05	0.0025
120	1	0.208333333

CSF Hs values	BW	TRI
	0.71125	0.791666

CSF	BW+TRI	F	F2
15	2	0.008333	6.94444E-05
14	2	0.008333	6.94444E-05
13	11	0.045833	0.002100694
12	78	0.325	0.105625
11	63	0.2625	0.06890625
10	56	0.233333	0.054444444
9	8	0.033333	0.001111111
8	14	0.058333	0.003402778
7	6	0.025	0.000625
	240	1	0.236354167

Ht= 0.763645
Hs= 0.751458
Fst= 0.015959

CFS	BB	F BB	F2 BB
15	0	0	0
14	2	0.016666	2.77778E-04
13	7	0.058333	0.003402778
12	33	0.275	0.075625
11	22	0.183333	0.033611111
10	37	0.308333	0.095069444
9	8	0.066666	0.004444444
8	6	0.05	0.0025
7	5	0.041666	0.001736111
	120	1	0.216666667

BAH	F BAH	F2 BAH
1	0.008333333	6.94444E-05
0	0	0
8	0.066666667	0.004444444
37	0.308333333	0.095069444
35	0.291666667	0.085069444
25	0.208333333	0.043402778
2	0.016666667	2.77778E-04
5	0.041666667	0.001736111
7	0.058333333	0.003402778
120	1	0.233472222

CSF Hs Values	BB	BAH
	0.783333	0.766527

CSF	BB+BAH	F	F2
15	1	0.004166	1.73611E-05
14	2	0.008333	6.94444E-05
13	15	0.0625	0.00390625
12	70	0.291666	0.085069444
11	57	0.2375	0.05640625
10	62	0.258333	0.066736111
9	10	0.041666	0.001736111
8	11	0.045833	0.002100694
7	12	0.05	0.0025
	240	1	0.218541667

Ht= 0.781458
Hs= 0.774930
Fst= 0.008353

CFS	BB	F BB	F2 BB
15	0	0	0
14	2	0.016666	2.77778E-04
13	7	0.058333	0.003402778
12	33	0.275	0.075625
11	22	0.183333	0.033611111
10	37	0.308333	0.095069444
9	8	0.066666	0.004444444
8	6	0.05	0.0025
7	5	0.041666	0.001736111
	120	1	0.216666667

JAM	F JAM	F2 JAM
0	0	0
1	0.008333333	6.94444E-05
9	0.075	0.005625
26	0.216666667	0.046944444
26	0.216666667	0.046944444
29	0.241666667	0.058402778
8	0.066666667	0.004444444
10	0.083333333	0.006944444
11	0.091666667	0.008402778
120	1	0.177777778

CSF Hs values	BB	JAM
	0.783333	0.822222

CSF	BB+JAM	F	F2
15	0	0	0
14	3	0.0125	0.00015625
13	16	0.066666	0.004444444
12	59	0.245833	0.060434028
11	48	0.2	0.04
10	66	0.275	0.075625
9	16	0.066666	0.004444444
8	16	0.066666	0.004444444
7	16	0.066666	0.004444444
	240	1	0.193993056

Ht= 0.806006
Hs= 0.802777
Fst= 0.004006

CSF	BB	F BB	F2 BB
15	0	0	0
14	2	0.016666	2.77778E-04
13	7	0.058333	0.003402778
12	33	0.275	0.075625
11	22	0.183333	0.033611111
10	37	0.308333	0.095069444
9	8	0.066666	0.004444444
8	6	0.05	0.0025
7	5	0.041666	0.001736111
	120	1	0.216666667

TRI	F TRI	F2 TRI
0	0	0
1	0.008333333	6.94444E-05
7	0.058333333	0.003402778
34	0.283333333	0.080277778
23	0.191666667	0.036736111
32	0.266666667	0.071111111
3	0.025	0.000625
14	0.116666667	0.013611111
6	0.05	0.0025
120	1	0.208333333

CSF Hs values	BB	TRI
	0.783333	0.791666

CSF	BB+TRI	F	F2
15	0	0	0
14	3	0.0125	0.00015625
13	14	0.058333	0.003402778
12	67	0.279166	0.077934028
11	45	0.1875	0.03515625
10	69	0.2875	0.08265625
9	11	0.045833	0.002100694
8	20	0.083333	0.006944444
7	11	0.045833	0.002100694
	240	1	0.210451389

Ht=	0.789548
Hs=	0.7875
Fst=	0.002594

CSF	BAH	F BAH	F2 BAH
15	1	0.008333	6.94444E-05
14	0	0	0
13	8	0.066666	0.004444444
12	37	0.308333	0.095069444
11	35	0.291666	0.085069444
10	25	0.208333	0.043402778
9	2	0.016666	2.77778E-04
8	5	0.041666	0.001736111
7	7	0.058333	0.003402778
	120	1	0.233472222

JAM	F JAM	F2 JAM
0	0	0
1	0.008333333	6.94444E-05
9	0.075	0.005625
26	0.216666667	0.046944444
26	0.216666667	0.046944444
29	0.241666667	0.058402778
8	0.066666667	0.004444444
10	0.083333333	0.006944444
11	0.091666667	0.008402778
120	1	0.177777778

CSF Hs values	BAH	JAM
	0.766527	0.822222

CSF	BAH+JAM	F	F2
15	1	0.004166	1.73611E-05
14	1	0.004166	1.73611E-05
13	17	0.070833	0.005017361
12	63	0.2625	0.06890625
11	61	0.254166	0.064600694
10	54	0.225	0.050625
9	10	0.041666	0.001736111
8	15	0.0625	0.00390625
7	18	0.075	0.005625
	240	1	0.200451389

Ht= 0.799548
Hs= 0.794375
Fst= 0.006470

CSF	BAH	F BAH	F2 BAH
15	1	0.008333	6.94444E-05
14	0	0	0
13	8	0.066666	0.004444444
12	37	0.308333	0.095069444
11	35	0.291666	0.085069444
10	25	0.208333	0.043402778
9	2	0.016666	2.77778E-04
8	5	0.041666	0.001736111
7	7	0.058333	0.003402778
	120	1	0.233472222

TRI	F TRI	F2 TRI
0	0	0
1	0.008333333	6.94444E-05
7	0.058333333	0.003402778
34	0.283333333	0.080277778
23	0.191666667	0.036736111
32	0.266666667	0.071111111
3	0.025	0.000625
14	0.116666667	0.013611111
6	0.05	0.0025
120	1	0.208333333

CSF Hs value	BAH	TRI
	0.766527	0.791666

CSF	BAH+TRI	F	F2
15	1	0.004166	1.73611E-05
14	1	0.004166	1.73611E-05
13	15	0.0625	0.00390625
12	71	0.295833	0.087517361
11	58	0.241666	0.058402778
10	57	0.2375	0.05640625
9	5	0.020833	4.34028E-04
8	19	0.079166	0.006267361
7	13	0.054166	0.002934028
	240	1	0.215902778

Ht= 0.784097
Hs= 0.779097
Fst= 0.006376

CSF	JAM	F JAM	F2 JAM
15	0	0	0
14	1	0.008333	6.94444E-05
13	9	0.075	0.005625
12	26	0.216666	0.046944444
11	26	0.216666	0.046944444
10	29	0.241666	0.058402778
9	8	0.066666	0.004444444
8	10	0.083333	0.006944444
7	11	0.091666	0.008402778
	120	1	0.177777778

TRI	F TRI	F2 TRI
0	0	0
1	0.008333333	6.94444E-05
7	0.058333333	0.003402778
34	0.283333333	0.080277778
23	0.191666667	0.036736111
32	0.266666667	0.071111111
3	0.025	0.000625
14	0.116666667	0.013611111
6	0.05	0.0025
120	1	0.208333333

CFS Hs Value	JAM	TRI
	0.822222	0.791666

CSF	JAM+TRI	F	F2
15	0	0	0
14	2	0.008333	6.94444E-05
13	16	0.066666	0.004444444
12	60	0.25	0.0625
11	49	0.204166	0.041684028
10	61	0.254166	0.064600694
9	11	0.045833	0.002100694
8	24	0.1	0.01
7	17	0.070833	0.005017361
	240	1	0.190416667

Ht= 0.809583
Hs= 0.806944
Fst= 0.003259

TPOX	BW	F BW	F2 BW
13	0	0	0
12	4	0.033333	0.001111111
11	30	0.25	0.0625
10	11	0.091666	0.008402778
9	16	0.133333	0.017777778
8	59	0.491666	0.241736111
7	0	0	0
6	0	0	0
	120	1	0.331527778

BB	F BB	F2 BB
0	0	0
5	0.041666667	0.001736111
25	0.208333333	0.043402778
13	0.108333333	0.011736111
20	0.166666667	0.027777778
44	0.366666667	0.134444444
7	0.058333333	0.003402778
6	0.05	0.0025
120	1	0.225

TPOX Hs value	BW	BB
	0.668472	0.775

TPOX	BW+BB	F	F2
13	0	0	0
12	9	0.0375	0.00140625
11	55	0.229166	0.052517361
10	24	0.1	0.01
9	36	0.15	0.0225
8	103	0.429166	0.184184028
7	7	0.029166	8.50694E-04
6	6	0.025	0.000625
	240	1	0.272083333

Ht= 0.727916
Hs= 0.721736
Fst= 0.008490

TPOX	BW	F BW	F2 BW
13	0	0	0
12	4	0.033333	0.001111111
11	30	0.25	0.0625
10	11	0.091666	0.008402778
9	16	0.133333	0.017777778
8	59	0.491666	0.241736111
7	0	0	0
6	0	0	0
	120	1	0.331527778

BAH	F BAH	F2 BAH
1	0.008333333	6.94444E-05
4	0.033333333	0.001111111
32	0.266666667	0.071111111
12	0.1	0.01
31	0.258333333	0.066736111
22	0.183333333	0.033611111
4	0.033333333	0.001111111
14	0.116666667	0.013611111
120	1	0.197361111

TPOX Hs Values	BW	BAH
	0.668472	0.802638

TPOX	BW+BAH	F	F2
13	1	0.004166	1.73611E-05
12	8	0.033333	0.001111111
11	62	0.258333	0.066736111
10	23	0.095833	0.009184028
9	47	0.195833	0.038350694
8	81	0.3375	0.11390625
7	4	0.016666	2.77778E-04
6	14	0.058333	0.003402778
	240	1	0.232986111

Ht= 0.767013
Hs= 0.735555
Fst= 0.041014

TPOX	BW	F BW	F2 BW
13	0	0	0
12	4	0.033333	0.001111111
11	30	0.25	0.0625
10	11	0.091666	0.008402778
9	16	0.133333	0.017777778
8	59	0.491666	0.241736111
7	0	0	0
6	0	0	0
	120	1	0.331527778

JAM	F JAM	F2 JAM
0	0	0
4	0.033333333	0.001111111
23	0.191666667	0.036736111
10	0.083333333	0.006944444
36	0.3	0.09
37	0.308333333	0.095069444
1	0.008333333	6.94444E-05
9	0.075	0.005625
120	1	0.235555556

TPOX Hs Values	BW	JAM
	0.668472	0.764444

TPOX	BW+JAM	F	F2
13	0	0	0
12	8	0.033333	0.001111111
11	53	0.220833	0.048767361
10	21	0.0875	0.00765625
9	52	0.216666	0.046944444
8	96	0.4	0.16
7	1	0.004166	1.73611E-05
6	9	0.0375	0.00140625
	240	1	0.265902778

Ht= 0.734097
Hs= 0.716458
Fst= 0.024028

TPOX	BW	F BW	F2 BW
13	0	0	0
12	4	0.033333	0.001111111
11	30	0.25	0.0625
10	11	0.091666	0.008402778
9	16	0.133333	0.017777778
8	59	0.491666	0.241736111
7	0	0	0
6	0	0	0
	120	1	0.331527778

TRI	F TRI	F2 TRI
1	0.008333333	6.94444E-05
6	0.05	0.0025
32	0.266666667	0.071111111
12	0.1	0.01
18	0.15	0.0225
41	0.341666667	0.116736111
2	0.016666667	2.77778E-04
8	0.066666667	0.004444444
	1	0.227638889

TPOX Hs values	BW	TRI
	0.668472	0.772361

TPOX	BW+TRI	F	F2
13	1	0.004166	1.73611E-05
12	10	0.041666	0.001736111
11	62	0.258333	0.066736111
10	23	0.095833	0.009184028
9	34	0.141666	0.020069444
8	100	0.416666	0.173611111
7	2	0.008333	6.94444E-05
6	8	0.033333	0.001111111
	240	1	0.272534722

Ht= 0.727465
Hs= 0.720416
Fst= 0.009689

TPOX	BB	F BB	F2 BB
13	0	0	0
12	5	0.041666	0.001736111
11	25	0.208333	0.043402778
10	13	0.108333	0.011736111
9	20	0.166666	0.027777778
8	44	0.366666	0.134444444
7	7	0.058333	0.003402778
6	6	0.05	0.0025
	120	1	0.225

BAH	F BAH	F2 BAH
1	0.008333333	6.94444E-05
4	0.033333333	0.001111111
32	0.266666667	0.071111111
12	0.1	0.01
31	0.258333333	0.066736111
22	0.183333333	0.033611111
4	0.033333333	0.001111111
14	0.116666667	0.013611111
120	1	0.197361111

TPOX Hs Value	BB	BAH
	0.775	0.802638

TPOX	BB+BAH	F	F2
13	1	0.004166	1.73611E-05
12	9	0.0375	0.00140625
11	57	0.2375	0.05640625
10	25	0.104166	0.010850694
9	51	0.2125	0.04515625
8	66	0.275	0.075625
7	11	0.045833	0.002100694
6	20	0.083333	0.006944444
	240	1	0.198506944

Ht= 0.801493
Hs= 0.788819
Fst= 0.015812

TPOX	BB	F BB	F2 BB
13	0	0	0
12	5	0.041666	0.001736111
11	25	0.208333	0.043402778
10	13	0.108333	0.011736111
9	20	0.166666	0.027777778
8	44	0.366666	0.134444444
7	7	0.058333	0.003402778
6	6	0.05	0.0025
	120	1	0.225

JAM	F JAM	F2 JAM
0	0	0
4	0.033333333	0.001111111
23	0.191666667	0.036736111
10	0.083333333	0.006944444
36	0.3	0.09
37	0.308333333	0.095069444
1	0.008333333	6.94444E-05
9	0.075	0.005625
120	1	0.235555556

TPOX Hs Values	BB	JAM
	0.775	0.764444

TPOX	BB+JAM	F	F2
13	0	0	0
12	9	0.0375	0.00140625
11	48	0.2	0.04
10	23	0.095833	0.009184028
9	56	0.233333	0.054444444
8	81	0.3375	0.11390625
7	8	0.033333	0.001111111
6	15	0.0625	0.00390625
	240	1	0.223958333

Ht= 0.776041
Hs= 0.769722
Fst= 0.008143

TPOX	BB	F BB	F2 BB
13	0	0	0
12	5	0.041666	0.001736111
11	25	0.208333	0.043402778
10	13	0.108333	0.011736111
9	20	0.166666	0.027777778
8	44	0.366666	0.134444444
7	7	0.058333	0.003402778
6	6	0.05	0.0025
	120	1	0.225

TRI	F TRI	F2 TRI
1	0.008333333	6.94444E-05
6	0.05	0.0025
32	0.266666667	0.071111111
12	0.1	0.01
18	0.15	0.0225
41	0.341666667	0.116736111
2	0.016666667	2.77778E-04
8	0.066666667	0.004444444
120	1	0.227638889

TPOX Hs Values	BB	TRI
	0.775	0.772361

TPOX	BB+TRI	F	F2
13	1	0.004166	1.73611E-05
12	11	0.045833	0.002100694
11	57	0.2375	0.05640625
10	25	0.104166	0.010850694
9	38	0.158333	0.025069444
8	85	0.354166	0.125434028
7	9	0.0375	0.00140625
6	14	0.058333	0.003402778
	240	1	0.2246875

Ht= 0.775312
Hs= 0.773680
Fst= 0.002104

TPOX	BAH	F BAH	F2 BAH
13	1	0.008333	6.94444E-05
12	4	0.033333	0.001111111
11	32	0.266666	0.071111111
10	12	0.1	0.01
9	31	0.258333	0.066736111
8	22	0.183333	0.033611111
7	4	0.033333	0.001111111
6	14	0.116666	0.013611111
	120	1	0.197361111

JAM	F JAM	F2 JAM
0	0	0
4	0.033333333	0.001111111
23	0.191666667	0.036736111
10	0.083333333	0.006944444
36	0.3	0.09
37	0.308333333	0.095069444
1	0.008333333	6.94444E-05
9	0.075	0.005625
120	1	0.235555556

TPOX Hs Value	BAH	JAM
	0.802638	0.764444

TPOX	BAH+JAM	F	F2
13	1	0.004166	1.73611E-05
12	8	0.033333	0.001111111
11	55	0.229166	0.052517361
10	22	0.091666	0.008402778
9	67	0.279166	0.077934028
8	59	0.245833	0.060434028
7	5	0.020833	4.34028E-04
6	23	0.095833	0.009184028
	240	1	0.210034722

Ht= 0.789965
Hs= 0.783541
Fst= 0.008131

TPOX	BAH	F BAH	F2 BAH
13	1	0.008333	6.94444E-05
12	4	0.033333	0.001111111
11	32	0.266666	0.071111111
10	12	0.1	0.01
9	31	0.258333	0.066736111
8	22	0.183333	0.033611111
7	4	0.033333	0.001111111
6	14	0.116666	0.013611111
	120	1	0.197361111

TRI	F TRI	F2 TRI
1	0.008333333	6.94444E-05
6	0.05	0.0025
32	0.266666667	0.071111111
12	0.1	0.01
18	0.15	0.0225
41	0.341666667	0.116736111
2	0.016666667	2.77778E-04
8	0.066666667	0.004444444
120	1	0.227638889

TPOX Hs Values	BAH	TRI
	0.802638	0.772361

TPOX	BAH+TRI	F	F2
13	2	0.008333	6.94444E-05
12	10	0.041666	0.001736111
11	64	0.266666	0.071111111
10	24	0.1	0.01
9	49	0.204166	0.041684028
8	63	0.2625	0.06890625
7	6	0.025	0.000625
6	22	0.091666	0.008402778
	240	1	0.202534722

Ht= 0.797465
Hs= 0.7875
Fst= 0.012496

TPOX	JAM	F JAM	F2 JAM
13	0	0	0
12	4	0.033333	0.001111111
11	23	0.191666	0.036736111
10	10	0.083333	0.006944444
9	36	0.3	0.09
8	37	0.308333	0.095069444
7	1	0.008333	6.94444E-05
6	9	0.075	0.005625
	120	1	0.235555556

TRI	F TRI	F2 TRI
1	0.008333333	6.94444E-05
6	0.05	0.0025
32	0.266666667	0.071111111
12	0.1	0.01
18	0.15	0.0225
41	0.341666667	0.116736111
2	0.016666667	2.77778E-04
8	0.066666667	0.004444444
120	1	0.227638889

TPOX Hs Values	JAM	TRI
	0.764444	0.772361

TPOX	JAM+TRI	F	F2
13	1	0.004166	1.73611E-05
12	10	0.041666	0.001736111
11	55	0.229166	0.052517361
10	22	0.091666	0.008402778
9	54	0.225	0.050625
8	78	0.325	0.105625
7	3	0.0125	0.00015625
6	17	0.070833	0.005017361
	240	1	0.224097222

Ht= 0.775902
Hs= 0.768402
Fst= 0.009666

THOI	BW	F BW	F2 BW
11	0	0	0
10	3	0.025	0.000625
9.3	28	0.233333	0.054444444
9	19	0.158333	0.025069444
8	17	0.141666	0.020069444
7	31	0.258333	0.066736111
6	22	0.183333	0.033611111
5	0	0	0
	120	1	0.200555556

BB	F BB	F2 BB
0	0	0
0	0	0
9	0.075	0.005625
18	0.15	0.0225
27	0.225	0.050625
47	0.391666667	0.153402778
19	0.158333333	0.025069444
0	0	0
120	1	0.257222222

THOI Hs Values	BW	BB
	0.799444	0.742777

THOI	BW+BB	F	F2
11	0	0	0
10	3	0.0125	0.00015625
9.3	37	0.154166	0.023767361
9	37	0.154166	0.023767361
8	44	0.183333	0.033611111
7	78	0.325	0.105625
6	41	0.170833	0.029184028
5	0	0	0
	240	1	0.216111111

Ht= 0.783888
Hs= 0.771111
Fst= 0.016300

THOI	BW	F BW	F2 BW
11	0	0	0
10	3	0.025	0.000625
9.3	28	0.233333	0.054444444
9	19	0.158333	0.025069444
8	17	0.141666	0.020069444
7	31	0.258333	0.066736111
6	22	0.183333	0.033611111
5	0	0	0
	120	1	0.200555556

BAH	F BAH	F2 BAH
0	0	0
2	0.016666667	2.77778E-04
10	0.083333333	0.006944444
17	0.141666667	0.020069444
31	0.258333333	0.066736111
48	0.4	0.16
12	0.1	0.01
0	0	0
120	1	0.264027778

THO1	BW	BAH
Hs	0.799444	0.735972

Values

THO1	BW+BAH	F	F2
11	0	0	0
10	5	0.020833	4.34028E-04
9.3	38	0.158333	0.025069444
9	36	0.15	0.0225
8	48	0.2	0.04
7	79	0.329166	0.108350694
6	34	0.141666	0.020069444
5	0	0	0
	240	1	0.216423611

Ht= 0.783576
Hs= 0.767708
Fst= 0.020250

THOI	BW	F BW	F2 BW
11	0	0	0
10	3	0.025	0.000625
9.3	28	0.233333	0.054444444
9	19	0.158333	0.025069444
8	17	0.141666	0.020069444
7	31	0.258333	0.066736111
6	22	0.183333	0.033611111
5	0	0	0
	120	1	0.200555556

JAM	F JAM	F2 JAM
0	0	0
2	0.016666667	2.77778E-04
4	0.033333333	0.001111111
14	0.116666667	0.013611111
27	0.225	0.050625
52	0.433333333	0.187777778
21	0.175	0.030625
0	0	0
120	1	0.284027778

THOI HsValues	BW	JAM
	0.799444	0.715972

THOI	BW+JAM	F	F2
11	0	0	0
10	5	0.020833	4.34028E-04
9.3	32	0.133333	0.017777778
9	33	0.1375	0.01890625
8	44	0.183333	0.033611111
7	83	0.345833	0.119600694
6	43	0.179166	0.032100694
5	0	0	0
	240	1	0.222430556

Ht= 0.777569
Hs= 0.757708
Fst= 0.025542

THOI	BW	F BW	F2 BW
11	0	0	0
10	3	0.025	0.000625
9.3	28	0.233333	0.054444444
9	19	0.158333	0.025069444
8	17	0.141666	0.020069444
7	31	0.258333	0.066736111
6	22	0.183333	0.033611111
5	0	0	0
	120	1	0.200555556

TRI	F TRI	F2 TRI
0	0	0
2	0.016666667	2.77778E-04
5	0.041666667	0.001736111
20	0.166666667	0.027777778
28	0.233333333	0.054444444
41	0.341666667	0.116736111
24	0.2	0.04
0	0	0
120	1	0.240972222

THOI Hs Values	BW	TRI
	0.799444	0.759027

THOI	BW+TRI	F	F2
11	0	0	0
10	5	0.020833	4.34028E-04
9.3	33	0.1375	0.01890625
9	39	0.1625	0.02640625
8	45	0.1875	0.03515625
7	72	0.3	0.09
6	46	0.191666	0.036736111
5	0	0	0
	240	1	0.207638889

Ht=	0.792361
Hs=	0.779236
Fst=	0.016564

THOI	BB	F BB	F2 BB
11	0	0	0
10	0	0	0
9.3	9	0.075	0.005625
9	18	0.15	0.0225
8	27	0.225	0.050625
7	47	0.391666	0.153402778
6	19	0.158333	0.025069444
5	0	0	0
	120	1	0.257222222

BAH	F BAH	F2 BAH
0	0	0
2	0.016666667	2.77778E-04
10	0.083333333	0.006944444
17	0.141666667	0.020069444
31	0.258333333	0.066736111
48	0.4	0.16
12	0.1	0.01
0	0	0
120	1	0.264027778

THOI HsValues	BB	BAH
	0.742777	0.735972

THOI	BB+BAH	F	F2
11	0	0	0
10	2	0.008333	6.94444E-05
9.3	19	0.079166	0.006267361
9	35	0.145833	0.021267361
8	58	0.241666	0.058402778
7	95	0.395833	0.156684028
6	31	0.129166	0.016684028
5	0	0	0
	240	1	0.259375

Ht=	0.740625
Hs=	0.739375
Fst=	0.001687

THOI	BB	F BB	F2 BB
11	0	0	0
10	0	0	0
9.3	9	0.075	0.005625
9	18	0.15	0.0225
8	27	0.225	0.050625
7	47	0.391666	0.153402778
6	19	0.158333	0.025069444
5	0	0	0
	120	1	0.257222222

JAM	F JAM	F2 JAM
0	0	0
2	0.016666667	2.77778E-04
4	0.033333333	0.001111111
14	0.116666667	0.013611111
27	0.225	0.050625
52	0.433333333	0.187777778
21	0.175	0.030625
0	0	0
120	1	0.284027778

THOI Hs Values	BB	JAM
	0.742777	0.715972

THOI	BB+JAM	F	F2
11	0	0	0
10	2	0.008333	6.94444E-05
9.3	13	0.054166	0.002934028
9	32	0.133333	0.017777778
8	54	0.225	0.050625
7	99	0.4125	0.17015625
6	40	0.166666	0.027777778
5	0	0	0
	240	1	0.269340278

Ht= 0.730659
Hs= 0.729375
Fst= 0.001758

THOI	BB	F BB	F2 BB
11	0	0	0
10	0	0	0
9.3	9	0.075	0.005625
9	18	0.15	0.0225
8	27	0.225	0.050625
7	47	0.391666	0.153402778
6	19	0.158333	0.025069444
5	0	0	0
	120	1	0.257222222

JAM	F JAM	F2 JAM
0	0	0
2	0.016666667	2.77778E-04
4	0.033333333	0.001111111
14	0.116666667	0.013611111
27	0.225	0.050625
52	0.433333333	0.187777778
21	0.175	0.030625
0	0	0
120	1	0.284027778

THOI Hs Values	BB	JAM
	0.742777	0.715972

THOI	BB+JAM	F	F2
11	0	0	0
10	2	0.008333	6.94444E-05
9.3	13	0.054166	0.002934028
9	32	0.133333	0.017777778
8	54	0.225	0.050625
7	99	0.4125	0.17015625
6	40	0.166666	0.027777778
5	0	0	0
	240	1	0.269340278

Ht=	0.730659
Hs=	0.729375
Fst=	0.001758

THOI	BB	F BB	F2 BB
11	0	0	0
10	0	0	0
9.3	9	0.075	0.005625
9	18	0.15	0.0225
8	27	0.225	0.050625
7	47	0.391666	0.153402778
6	19	0.158333	0.025069444
5	0	0	0
	120	1	0.257222222

TRI	F TRI	F2 TRI
0	0	0
2	0.016666667	2.77778E-04
5	0.041666667	0.001736111
20	0.166666667	0.027777778
28	0.233333333	0.054444444
41	0.341666667	0.116736111
24	0.2	0.04
0	0	0
120	1	0.240972222

THOI Hs Values	BB	TRI
	0.742777	0.759027

THOI	BB+TRI	F	F2
11	0	0	0
10	2	0.008333	6.94444E-05
9.3	14	0.058333	0.003402778
9	38	0.158333	0.025069444
8	55	0.229166	0.052517361
7	88	0.366666	0.134444444
6	43	0.179166	0.032100694
5	0	0	0
	240	1	0.247604167

Ht=	0.752395
Hs=	0.750902
Fst=	0.001984

THOI	BAH	F BAH	F2 BAH
11	0	0	0
10	2	0.016666	2.77778E-04
9.3	10	0.083333	0.006944444
9	17	0.141666	0.020069444
8	31	0.258333	0.066736111
7	48	0.4	0.16
6	12	0.1	0.01
5	0	0	0
	120	1	0.264027778

JAM	F JAM	F2 JAM
0	0	0
2	0.016666667	2.77778E-04
4	0.033333333	0.001111111
14	0.116666667	0.013611111
27	0.225	0.050625
52	0.433333333	0.187777778
21	0.175	0.030625
0	0	0
120	1	0.284027778

THOI Hs Values	BAH	JAM
	0.735972	0.715972

THOI	BAH+JAM	F	F2
11	0	0	0
10	4	0.016666	2.77778E-04
9.3	14	0.058333	0.003402778
9	31	0.129166	0.016684028
8	58	0.241666	0.058402778
7	100	0.416666	0.173611111
6	33	0.1375	0.01890625
5	0	0	0
	240	1	0.271284722

Ht= 0.728715
Hs= 0.725972
Fst= 0.003764

THOI	BAH	F BAH	F2 BAH
11	0	0	0
10	2	0.016666	2.77778E-04
9.3	10	0.083333	0.006944444
9	17	0.141666	0.020069444
8	31	0.258333	0.066736111
7	48	0.4	0.16
6	12	0.1	0.01
5	0	0	0
	120	1	0.264027778

TRI	F TRI	F2 TRI
0	0	0
2	0.016666667	2.77778E-04
5	0.041666667	0.001736111
20	0.166666667	0.027777778
28	0.233333333	0.054444444
41	0.341666667	0.116736111
24	0.2	0.04
0	0	0
120	1	0.240972222

THOI Hs	BAH	TRI
Values		
	0.735972	0.759027

THOI	BAH+TRI	F	F2
11	0	0	0
10	4	0.016666	2.77778E-04
9.3	15	0.0625	0.00390625
9	37	0.154166	0.023767361
8	59	0.245833	0.060434028
7	89	0.370833	0.137517361
6	36	0.15	0.0225
5	0	0	0
	240	1	0.248402778

Ht=	0.751597
Hs=	0.7475
Fst=	0.005451

THOI	JAM	F JAM	F2 JAM
11	0	0	0
10	2	0.016666	2.77778E-04
9.3	4	0.033333	0.001111111
9	14	0.116666	0.013611111
8	27	0.225	0.050625
7	52	0.433333	0.187777778
6	21	0.175	0.030625
5	0	0	0
	120	1	0.284027778

TRI	F TRI	F2 TRI
0	0	0
2	0.016666667	2.77778E-04
5	0.041666667	0.001736111
20	0.166666667	0.027777778
28	0.233333333	0.054444444
41	0.341666667	0.116736111
24	0.2	0.04
0	0	0
120	1	0.240972222

THOI Hs Values	JAM	TRI
	0.715972	0.759027

THOI	JAM+TRI	F	F2
11	0	0	0
10	4	0.016666	2.77778E-04
9.3	9	0.0375	0.00140625
9	34	0.141666	0.020069444
8	55	0.229166	0.052517361
7	93	0.3875	0.15015625
6	45	0.1875	0.03515625
5	0	0	0
	240	1	0.259583333

Ht= 0.740416
Hs= 0.7375
Fst= 0.003939

APPENDIX XI

G Test Values for CSF

	BB	BAH	JAM	TRI
BW	24.4116*	16.9111*	33.6854*	30.3304*
BB		12.6070	5.9669	6.2964
BAH			11.7592	10.0759
JAM				6.0579

G Test Values for TPOX

	BB	BAH	JAM	TRI
BW	16.3612*	40.7965*	23.7061*	14.8656*
BB		15.7348	10.8610	5.2652
BAH			9.7282	11.8822
JAM				9.6518

G Test Values for THO1

	BB	BAH	JAM	TRI
BW	18.5581*	19.5202*	26.5668*	20.4207*
BB		3.9482	4.7756	4.25686
BAH			5.7522	6.6130
JAM				2.6892

TABLE 2 : Above tables give G-test values for all three loci and possible combinations. The 2-way R x C contingency table calculating the G-statistic was carried out with a program provided by G. Carmody (Ottawa, Canada).
*probability values less than 0.01

APPENDIX XII

X^2 Values for CSF

	BB	BAH	JAM	TRI
BW	29.5593*	22.0087*	42.7025*	38.9094*
BB		14.0636	6.0456	6.4802
BAH			12.8414	11.0459
JAM				6.1724

X^2 Values for TPOX

	BB	BAH	JAM	TRI
BW	21.3928*	48.8640*	27.8186*	19.1355*
BB		16.3862	11.4945	5.8191
BAH			10.2998	12.0506
JAM				10.1708

X^2 Values FOR THO1

	BB	BAH	JAM	TRI
BW	20.2422*	20.0058*	28.8975*	22.0998*
BB		4.7346	5.6001	5.0470
BAH			5.8694	6.7239
JAM				2.6981

TABLE 3: χ^2 values for all loci with all possible population combinations.
*probability values less than 0.01

APPENDIX XIII

LOCI	T-VALUE	CRITICAL T-VALUE
CSF1PO	7.40*	2.306
TPOX	1.75	2.306
THO1	3.40*	2.306

TABLE 4: T-test values between Broward whites and African Descent population.* marks significant values.

APPENDIX XIV

LOCI	T-VALUE	CRITICAL T-VALUE
CSF1PO	-.2000*	3.182
TPOX	-.3256*	3.182
THO1	-.5862*	3.182

TABLE 5: T-values between Broward blacks and other African descent populations. * mark significant values.