Instrumentation and Materials needed:

Materials

- Dog volunteers
- Cytology brushes (Fisher Scientific, catalog number 892030-P25)
- Primers (We have used Integrated DNA Technologies, http://www.idtdna.com)
- Genomic DNA extraction kit (Recommended - QIAamp DNA mini kit, QIAGEN, catalog number 51304)
- 1 X PBS (400 uL per dog sample)
- PCR reagents (Recommended – Taq PCR master mix kit, QIAGEN, catalog number 201443)
- Restriction enzymes - These are specific to the dCAPs primers you are using. The seven primer sets described use BamHI, EcoRI, PstI, and SalI. New England Biolabs (www.neb.com) will provide classroom catalogs upon request. The catalogs can be used by students to identify conditions for their specific enzymes, and are a useful resource for learning about many molecular reagents.
- 1 X TBE buffer
- Loading Dye
- 100 bp ladder (NEB, catalog number N3231S)
- Ethidium Bromide or Gel Red (VWR, catalog number 89139-138)

Instruments

- Thermocycler
- Water baths
- Gel electrophoresis systems
- Gel documentation system
- (Recommended) Spectrophotometer for genomic DNA quantification
- (Optional) Computers for dCAPs primer design

Recipes for Reagents

1 X PBS:

8 g	NaCl
0.2 g	KCl
1.44 g	Na_2HPO_4
0.24 g	KH_2PO_4

Dissolve in 800 mL of distilled water. Adjust the pH to 7.4 with HCl, and then the final volume to 1 liter. Sterilize by autoclaving and store at room temperature.

10 X TBE buffer:

108 g	Tris base
55 g	Boric acid
40 mL	0.5M EDTA (pH 8.0)

Dissolve in 800 mL of distilled water. Adjust the final volume to 1 liter. Sterilize by autoclaving and store at room temperature. Dilute 1/10 with distilled water for a 1 X TBE working solution.

5 X Orange G gel loading dye:

7.5 ml glycerol
100 mg Orange G dye

Add dH2O to a final volume of 50 ml and sterile filter, store at 4^0C.