

## PCR: Duane (amplifluor)

Amplifluor: Yes

### Primers

Nucleotide sequence (5' --> 3')

Primer	Sequence
Duane-1	GAAGGTGACCAAGTTCATGCTCATATAAGCGGTTCCCTGTGGT
Duane-2	GAAGGTCGGAGTCAACGGATTCATATAAGCGGTTCCCTGTGGA
Duane-R	GTGTCTGGTCGATACTTGACAA

### Reagents

Reagents/Constituents	Commercial name	Stock Concentration	20x Primer mix concentration (μM)	Volume (μL)
DNA Sample				1.0
10x Buffer	10x Reaction Mix S Plus			0.5
dNTPs		2.5 mM		0.4
20x amplifluor SNP FAM Primer				
20x amplifluor SNP JOE Primer				
Primer: <b>Duane-1</b>			0.5	
Primer: <b>Duane-2</b>			0.5	
Primer: <b>Duane-R</b>			7.5	
Amplifluor primer volume				1.0
Water				1.58
Taq Polymerase	Platinum Taq (5U/l)			0.05

**Comments on protocol:** DNA Sample (cleaned ear punch in dH2O-see cleanup protocol).

Reactions to be done at room temperature with reagents added in order.

### Strategy

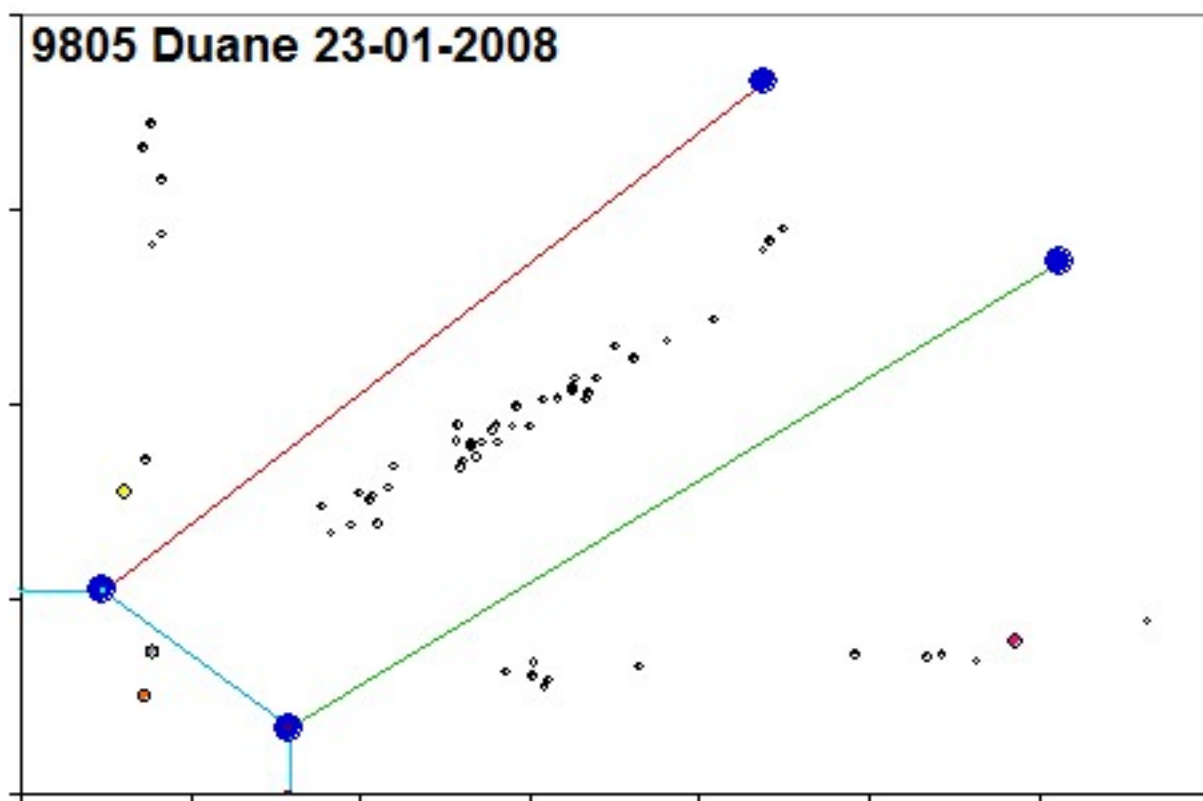
Steps	Temp (°C)	Time (seconds)
Hot start: <b>Yes</b>		
Initiation/Melting	94.0	240
Denaturation	94.0	15
Annealing	55.0	20
Elongation	72.0	30
Number of cycles (repeat denature, anneal & elongate): <b>35</b>		
Strand completion (i.e. 72°C, 10 min)	72.0	180
Finish (i.e. 4°C, indefinite)	10.0	Hold

### Electrophoresis

<b>Running buffer:</b>	
<b>% Agarose:</b>	
<b>Volt:</b>	
<b>Estimated running time (min):</b>	

### Image

9805 Duane 23-01-2008



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