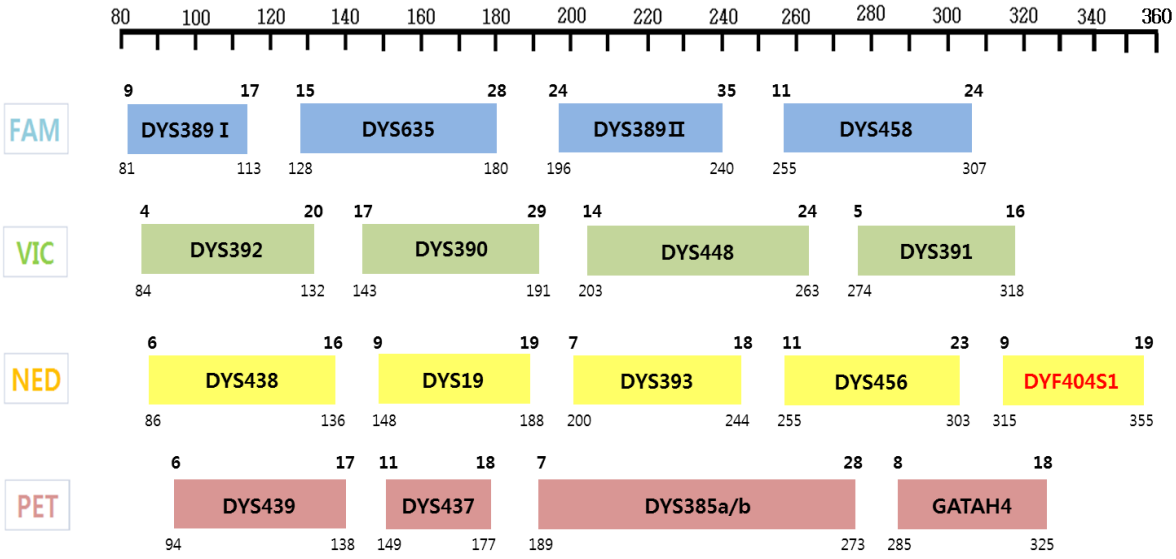
Kplex-Y18 PCR System Protocol

Allelic size range for 18 Y-STR loci:



5 X Primer Mix for Multiplex PCR:

STR loci		Primer Sequence (5'→3')	Conc. (μM)
DYS389I/II	F	FAM-CCA ACT CTC ATC TGT ATT ATC T	5.00
	R	GAT AGA TTG ATA GAG GGA GGG A	5.00
DYS635	F	TGG CTT CTC ACT TTG CAT AGA A	1.05
	R	FAM-ACC AGC CCA AAT ATC CAT CA	1.05
DYS458	F	FAM-GCA ACA GGA ATG AAA CTC CAA	2.00
	R	GAA GTA GCT GGG GCT AGA GGT T	2.00
DYS392	F	GTA AAC CTA CCA ATC CCA TTC CT	5.00
	R	VIC-AAA AGC CAA GAA GGA AAA CAA A	5.00
DYS390	F	GCA ATG TGT ATA CTC AGA AAC AAG G	2.00
	R	VIC-CTG CAT TTT GGT ACC CCA TA	2.00
DYS448	F	GCA GAA AGG GAG ATA GAG ACA TGG	1.40
	R	VIC-CCT CAT ATT TCT GGC CGG TCT	1.40
DYS391	F	VIC-CAA GAC ACC CCA CCA CAG AT	2.50
	R	GGC AAG CAA TTG CCA TAG AGG	2.50

Kplex-Y18 PCR System Protocol (Continued)

5 X Primer Mix for Multiplex PCR (Continued):

STR loci		Primer Sequence (5'→3')	Conc. (μM)
DYS438	F	NED-TGG GGA ATA GTT GAA CGG TAA	1.40
	R	GCA ACA AGA GTG AAA CTC CAT T	1.40
DYS19	F	NED-GGA GTC CAT CTG GGT TAA GGA	1.50
	R	CTA CTG AGT TTC TGT TAT AGT GTT TTT T	1.50
DYS393	F	TGT CTT TAC TAG CAG CAT GAG AAC	5.00
	R	NED-AAA CTC AAG TCC AAA AAA TGA GG	5.00
DYS456	F	GAC CCA GCC TAC ATC TTT CTC CA	4.25
	R	NED-CCA TCA ACT CAG CCC AAA AC	4.25
DYF404S1	F	NED-CCT GGT AAT CTT CCA AAA TCG T	3.25
	R	GAA AGA TCA AAG GAG CCC AG	3.25
DYS439	F	GGT GGA GAC AGA TAG ATG ATA AA	1.00
	R	PET-GGC TTG GAA TTC TTT TAC CCA	1.00
DYS437	F	PET-GAC TAT GGG CGT GAG TGC AT	1.80
	R	AGA TAG ATA GAT AAC CAC AGA TAA ATA T	1.80
DYS385ab	F	PET-GAA ATG AAA TTC AGA AAG GAA GG	5.00
	R	TTC CAA TTA CAT AGT CCT CCT TTC T	5.00
YGATAH4	F	TAA ACA ACT TAA CAG GAT AAA TCA CC	5.00
	R	PET-CAG AGT GGG TTC TGA AGA GCT A	5.00

Reagents Needed:

- 5 X Primer Mix for Kplex-Y18 system
- Gold ST*R 10 X Buffer (Promega, Madison, WI)
- AmpliTaq Gold® DNA Polymerase (Applied Biosystems, Foster City, CA)

PCR Mixture:

PCR Component	Vol. (μL)
dH_2O	5.4
5 X Primer Mix	2.0
10 X Gold ST*R Buffer	1.0
AmpliTaq Gold (5 U/μL)	0.6
Template DNA (1ng/μL)	1.0
Total	10.0

Thermal Cycling:

95℃ for 11 minutes, then:

94℃ for 20 seconds
60℃ for 60 seconds
72℃ for 45 seconds
for 29 cycles, then:

60℃ for 45 minutes
4℃ soak

Kplex-Y18 PCR System Protocol (Continued)

Capillary Electrophoresis

Materials and Reagents Needed:

- Dry heating block or thermal cycler
- 3130 capillary array, 36 cm (Applied Biosystems, Foster City, CA)
- Performance Optimized Polymer (POP4, Applied Biosystems, Foster City, CA)
- GeneScan™ 500 LIZ™ Size Standard
- Hi-Di™ Formamide (Applied Biosystems, Foster City, CA)

Mixture for CE run:

GeneScan™ 500 LIZ™ Size Standard	0.2 µL
Hi-Di™ Formamide	10.0 µL
PCR product	1.0 µL

Denaturation:

95℃ for 5 minutes
4℃ soak

3130 Data Collection Software:

Application type	HID
Injection voltage	3.0kV
Injection time	5 seconds
Run time	23 minutes

Download a PDF copy of this protocol:

http://forensic.yonsei.ac.kr/protocols.html

Genotyping result of 2800M control DNA