In situ Hybridization Data Sheet

Date: 18-3-2010

= Same as FISH-H

Name: Niaz

" FISH – K "

Cross-reference to notes or photographs of slides or materials.

Master-mix:

Component	Final concentration in hybridization mix	Amount for 1 slide	Multiply by number of slides +1
100% Formamide	50%	20 µl	
20x SSC	2x	4 µl	
50% Dextran sulphate	10%	8 µl	
Salmon sperm DNA 1µg/µl	0.025µg	1 µl	
100mM EDTA	1.25 mM	0.5µl	
10% SDS	0.125%	0.5µl	
Total		34µl	

Component	Typical value per slide	No 1 17/3/10 5156 B+	good Hy (CS13) 2 8/3/10 5109-R aa	? 3 10/3/10 5003 a	good ✓ 4 11/3/11 5019-S a	✓ 5 3/3/10 Reliant aa	Poor CS13 6 10/3/10 5078 BB+	Thin 7 5163 A	8 5121 a
Master-mix	34 µl								
Probe 1 CS-13 (Dig)	25-200ng					3.5µl ✓	3.5µl ✓		
Probe 2 2pTa1 - Bio	25-200ng	1.8µl ✓							
Probe 3 PTa794-Dig	25-200ng		3.5µl ✓		3.5µl ✓				
Probe 4 PTa-71 – Bio	25-200ng					0.6µl ✓			0.6µl ✓
Probe 5 pTa1-Dig	25-200ng			2.5µl ✓					
Probe 6 Afa-Thin (Bio) *1	25-200ng							1.5µl ✓	
Probe 7 Thin-DNA (Dig)	25-200ng	2.5µl ✓						2.5µl ✓	2.5µl ✓
Probe 8 *2 Thin-DNA (Bio)	25-200ng		1.6µl ✓	1.6µl ✓	1.6µl ✓		1.6µl ✓		
Probe 9	25-200ng								
Probe 10	25-200ng								
Blocking DNA	10-100x of probe	2.5µl ✓	2.5µl ✓	2.5µl	2.5µl	–	2.5µl	2.5µl ✓	2.5µl
Water	(to 40µl)								
Total Volume	40 µl								

*1 = Afa-Thin Bio = Randomly labelled 6/1/09

*2 = Thin-DNA -Bio = (Genomic — 15.1.9) Niaz

In situ Hybridization Data Sheet

Date: 20-4-2010

FISH – "L"

Same as FISH "H"

Name: Niaz

Cross-reference to notes or photographs of slides or materials.

Master-mix:

Component	Final concentration in hybridization mix	Amount for 1 slide x 22	Multiply by number of slides +1
100% Formamide	50%	20 µl	440µl
20x SSC	2x	4 µl	88µl
50% Dextran sulphate	10%	8 µl	176µl
Salmon sperm DNA 1µg/µl	0.025µg	1 µl	22µl
100mM EDTA	1.25 mM	0.5µl	11µl
10% SDS	0.125%	0.5µl	11µl
Total		34µl	Total = 748

Component	Typical value per slide	a 1 8/10 5109-R	A+ 2 8/3/10 5109-R	a+ 3 16/4/10 5078	aa 4 9/4/10 5121	B+ 5 20/4/10 5156	6 5075	Thin 7 Reliant	A+ 8 11/9/06 2016
Master-mix	34 µl				Date 15/4/10				
Probe 1 dpTa1-Bio	25-200ng	2.5µl			2.5µl				
Probe 2 CS-13-Bio	25-200ng			3µl			2.5µl		
Probe 3 CS-13-Dig	25-200ng		3µl					3µl	3.5µl
Probe 4 pTa71-Bio	25-200ng							0.6µl	
Probe 5 Afa-Thin-Bio	25-200ng								
Probe 6 Thin-DNA Bio	25-200ng		1.5µl			1.5µl			1.5µl
Probe 7 Thin DNA-Dig	25-200ng	2.5µl		2.5µl	2.5µl		2.5µl		
Probe 8 pTa71-Dig	25-200ng					1.3µl			
Probe 9	25-200ng								
Probe 10	25-200ng								
Blocking DNA	10-100x of probe	2.5µl	2.5µl	2.5µl	2.5µl	2.5µl	2.5µl	–	2.5µl
Water	(to 40µl)								
Total Volume	40 µl								

In situ Hybridization Data Sheet

Date: 14/10/2010

Name: Niaz "FISH-N"

Cross-reference to notes or photographs of slides or materials.

Master-mix:

Component	Final concentration in hybridization mix	Amount for 1 slide	Multiply by number of slides +1
100% Formamide	50%	20 µl	
20x SSC	2x	4 µl	
50% Dextran sulphate	10%	8 µl	
Salmon sperm DNA 1µg/µl	0.025µg	1 µl	
100mM EDTA	1.25 mM	0.5µl	
10% SDS	0.125%	0.5µl	
Total		34µl	

Component	Typical value per slide	1 (5) 5117 Aa 16-11-08	2 (5) 2016 A+ 11-9-08	3 5109 aa old	4 (yellow) KS95H102 aa+ 9/12/09	5 KS95H102 aa 9/12/09	6 KS96Hw10-1 Bb+ 22-12-09	7 (++) Lamberto 30-9-2	8 + Manaska aa 8/10/2010
Master-mix	34 µl								
Probe 1 *1 Pet-5 (Dig)	25-200ng						3µl	3µl	3µl
Probe 2 *2 CS-13 (Dig)	25-200ng	3µl							
Probe 3 *2 dpTa1-Bio	25-200ng			2.5µl					
Probe 4 pTa71-Bio	25-200ng								0.6µl
Probe 5 Thin-DNA (Dig)	25-200ng		2.5µl	2.5µl	2.5µl				
Probe 6 Thin-DNA (Bio)	25-200ng	1.8µl				1.8µl	1.8µl		
Probe 7 Rye-Bio	25-200ng							3µl	
Probe 8 *4 CS-13-I (Bio)	25-200ng		3µl						
Probe 9 *5 CS-13-II (Bio)	25-200ng				3µl				
Probe 10 *6 *5 CS-13-II-Dig	25-200ng					3µl			
Blocking DNA	10-100x of probe	2µl	2µl	2µl	2µl	2µl	2µl	2µl	–
Water	(to 40µl)								
Total Volume	40 µl								

*1 = PCR labelled – 30/9/2010
*2 = PCR-labelled – 30/9/2010
*3 = PCR-labelled – 30/9/10
*4 = Rand. Pr. – 30/9/10
*5 = Random Primed — *6 — [illegible]

5

In situ Hybridization Data Sheet

FISH – "O"

Date: 12-11-2010.

Name: Niaz

Cross-reference to notes or photographs of slides or materials. Same as "fish - H"

Master-mix:

Component	Final concentration in hybridization mix	Amount for 1 slide	Multiply by number of slides +1
100% Formamide	50%	20 µl	✓ ✓
20x SSC	2x	4 µl	48 ✓ ✓
50% Dextran sulphate	10%	8 µl	96 ✓
Salmon sperm DNA 1µg/µl	0.025µg	1 µl	12 ✓ ✓
100mM EDTA	1.25 mM	0.5µl	6 ✓
10% SDS	0.125%	0.5µl	6 ✓
Total		34µl	

Component	Typical value per slide	1 +/+ KS96H010-1 a 6/11/2010	2 KS96H010-1 BB+	3 KS95H102 aa 9/12/09	4 ✓ +/? 5057 aa 9/12/09	5 5057 aa+ 9/12/09	6 +/+ ✓ yellow 5106 a 3-3-10	7 blue 5/06 a 3-3-10	8 5109-R+ a 15-10-10
Master-mix	34 µl								
Probe 1 Thin-DNA Bio	25-200ng	3µl		2.5µl		3µl	2.5µl		2.5µl
Probe 2 Thin DNA Dig	25-200ng		2.5µl		2.5µl			2.5µl	
Probe 3 Pet-5-Dig	25-200ng	4µl		~~4µl~~ 3.5µl		3µl	4µl		4µl
Probe 4 CS-13-I (Bio)	25-200ng		4µl		4µl			4µl	
Probe 5	25-200ng								
Probe 6	25-200ng								
Probe 7	25-200ng								
Probe 8	25-200ng								
Probe 9	25-200ng								
Probe 10	25-200ng								
Blocking DNA	10-100x of probe	3.5µl	3µl	3µl	3µl	3.5µl	3µl	3µl	3µl
Water	(to 40µl)								
Total Volume	40 µl								

→ All repet. probes that worked in "FISH - N" are used here.

– by mistake i used wrong probes for fish "6" and the [illegible]