

## General info

### Protocol information

Protocol name	SAX_SP3_KF2_v2
Modified by	chris
Kit name	KF2
Description	KingFisher Flex 96 protocol for precipitation and digestion of captured plasma particles ending with tryptic peptides. This is Part 2 of 2. Chris Wu, MacCoss Laboratory, University of Washington

## Sample layout

## Reagent info






P1 KF1 Plate 7		96 DW plate	
Name	Well volume [µl]	Total reagent volume [µl]	Type
100% ACN	350	-	Reagent
Plasma-SDS-Tris-TCEP- IAA-DTT	100	-	Sample
Beads	25	-	Reagent
P2 ACN Wash 1		96 DW plate	
Name	Well volume [µl]	Total reagent volume [µl]	Type
95% Acetonitrile	1000	-	Reagent
P3 ACN Wash 2		96 DW plate	
Name	Well volume [µl]	Total reagent volume [µl]	Type
95% Acetonitrile	1000	-	Reagent
P4 ACN Wash 3		96 DW plate	
Name	Well volume [µl]	Total reagent volume [µl]	Type
95% Acetonitrile	1000	-	Reagent
P5 EtOH Wash 1		96 DW plate	
Name	Well volume [µl]	Total reagent volume [µl]	Type
70% Ethanol	1000	-	Reagent
P6 EtOH Wash 2		96 DW plate	
Name	Well volume [µl]	Total reagent volume [µl]	Type
70% Ethanol	1000	-	Reagent
P7 Elution - Trypsin		96 DW plate	
Name	Well volume [µl]	Total reagent volume [µl]	Type
50mM Tris, Trypsin 1:20	200	-	Reagent
Tip Plate		96 standard plate	
Name	Well volume [µl]	Total reagent volume [µl]	Type
-	-	-	-

## Dispensed reagents

The protocol does not contain dispensed reagents

## Steps data

	Tip1	96 DW tip comb	
	Pick-Up	Tip Plate	
	Bind Proteins	P1 KF1 Plate 7	
	Beginning of step	Precollect	No
		Release beads	No
	Mixing / heating:	Shake 1 time, speed	00:01:00, Medium
		Shake 2 time, speed	00:10:00, Fast
		Loop count	2
		Heating during mixing	No
	End of step	Postmix	No
		Collect count	5
		Collect time [s]	30
	Wash 1	P2 ACN Wash 1	
	Beginning of step	Precollect	No
		Release beads	No
	Mixing / heating:	Mixing time, speed	00:02:30, Slow
		Heating during mixing	No
	End of step	Postmix	No
		Collect beads	No
	Wash 2	P3 ACN Wash 2	
	Beginning of step	Precollect	No
		Release beads	No
	Mixing / heating:	Mixing time, speed	00:02:30, Slow
		Heating during mixing	No
	End of step	Postmix	No
		Collect beads	No
	Wash 3	P4 ACN Wash 3	
	Beginning of step	Precollect	No
		Release beads	No
	Mixing / heating:	Mixing time, speed	00:02:30, Slow
		Heating during mixing	No
	End of step	Postmix	No
		Collect beads	No
	Wash 4	P5 EtOH Wash 1	
	Beginning of step	Precollect	No
		Release beads	No
	Mixing / heating:	Mixing time, speed	00:02:30, Slow
		Heating during mixing	No
	End of step	Postmix	No
		Collect beads	No

	Wash 5	P6 EtOH Wash 2	
	Beginning of step	Precollect	No
		Release beads	No
	Mixing / heating:	Mixing time, speed	00:02:30, Slow
		Heating during mixing	No
	End of step	Postmix	No
		Collect beads	No
	Pause1	P7 Elution - Trypsin	
		Message	Add digestion/elution plate
	Protein Digestion and Elution	P7 Elution - Trypsin	
	Beginning of step	Precollect	No
		Release time, speed	00:00:20, Bottom mix
	Mixing / heating:	Shake 1 time, speed	00:00:15, Medium
		Shake 2 time, speed	00:02:15, Paused
		Loop count	27
		Tip position when paused	Tip edge in liquid
		Heating temperature [°C]	47
		Preheat	No
	End of step	Postmix	No
		Collect count	5
		Collect time [s]	30
	Dispose Beads	P1 KF1 Plate 7	
		Release time, speed	00:00:30, Medium
	Leave	Tip Plate	

## Lot info

No lot numbers have been defined.