Clustering Sheet for Flowcell **HVCFWDSX7**

Name
Bartlomiej Gebarski

Sequencer
NovaSeq1A

Run Type
NovaSeq S4 PE150 (r1: 150, r2: 150)

Clustering Date
2023-12-05

Sequencing Date
2023-12-05

Status
Analyzed

1st Index Length
15

2nd Index Length
15

Comments
no comment

Reagents Checked Out
☐

- NovaSeq Xp 4-Lane Kit: 1

- NovaSeq 6000 S4 Reagent Kit (300 cycles): 1

num	Id	Scientist	Custom	PR	Comments
1	⊎M15213_R14052_1	Volodymyr Shubchynskyy	☐	Standard	Solexa-Control
	⊎M15894_R14721_1	Volodymyr Shubchynskyy			Solexa-Control
	⊎M15899_R14726_1	Volodymyr Shubchynskyy			Solexa-Control
	⊎M16108_R14923_1	Volodymyr Shubchynskyy			Solexa-Control
	⊎M16448_R15221_1	Philip Wolff			Solexa-Control
	⊎M16872_R15617_1	Volodymyr Shubchynskyy			Solexa-Control
	M17615_R16301_1	Fernando Becerril Perez			BRB/FS-seq
	M17621_R16307_1	Sophie Bauer			RNA-Seq
	⊎M17625_R16309_1	Dagmar Gotthardt			Bulk Smart-Seq 3 RNA-Seq
	M17675_R16204_2	Abel Vertesy			[various]
	⊎M17711_R16396_1	Volodymyr Shubchynskyy			Solexa-Control
	⊎M17724_R16408_1	Sandor Urmosi-Incze			Solexa-Control
	⊎M17733_R16420_1	Sandor Urmosi-Incze			Solexa-Control
	⊎M17742_R16429_1	Jeanne Fesselet			SLAM-Seq
	⊎M17754_R16439_1	Sandor Urmosi-Incze			Solexa-Control
	⊎268806_R16398_1	Logan Hodgskiss			Small RNA seq
2	M17675_R16204_1	Abel Vertesy	☐	Standard	[various]
3	M17562_R16248_1	Ulla Schellhaas	☐	Standard	SLAM-seq
4	M17687_R16371_1	Roland Zauner	☐	Standard	10x RNA

Instructions

1. Check, if instrument is idle. If run just finished - press 'Home' button and prepare new run.
2. Plan ahead every run, reagents can be thawn over night in cold room or in water for 2-4h. After thawing in cold room it is important to check if everything was thawn.
3. Prepare Cluster Sheet, Sample and all reagents.
4. Take out the flowcell from the fridge to equilibrate it to room temperature (5-10min).
5. Denature the samples in Eppendorf tubes:
   - Lane 1: M15213, M15894, M15899, M16108, M16448, M16872, M17615, M17621, M17625, M17675, M17711, M17724, M17733, M17742, M17754, 268806
     - **18.84µl** of buffer
     - **8.91µl** of **2.5nM** library;
     - Add **2.25µl** of **0.1nM** PhiX;
   - Lane 2: M17675
     - **16.61µl** of buffer
     - **11.14µl** of **2nM** library;
     - Add **2.25µl** of **0.1nM** PhiX;
   - Lane 3: M17562
     - **18.84µl** of buffer
     - **8.91µl** of **2.5nM** library;
     - Add **2.25µl** of **0.1nM** PhiX;
   - Lane 4: M17687
     - **18.84µl** of buffer
     - **8.91µl** of **2.5nM** library;
     - Add **2.25µl** of **0.1nM** PhiX;
   - Add **7** µl of **0.2N** NaOH to each sample;
   - Incubate for 8 minutes (Take out DPX set from the freezer to thaw, up to 10 minutes);
   - During incubation install flowcell and gasket in the holder;
   - Neutralize by adding **8** µl of **400mM** Tris;
6. Put denatured samples on ice and prepare ExAmp mix:
   - **315 µl** of DPX1
   - **45 µl** of DPX2
   - **165 µl** of DPX3
   - Vortex and spin down;
7. Transfer **105µl** of mix to each sample;
8. Vortex and spin down;
9. Load **130µl** of final mix in each well;
10. Start sequencing within 30 minutes, follow instructions on the screen.