1. **Purpose**

This procedure provides instructions for the maintenance of the Illumina MiSeq to ensure the equipment functions according to established criteria to produce the quality of products and services required by the *“insert laboratory name here”*.

1. **Scope**

This document applies to Illumina MiSeqused within the *(Your Lab / Branch, etc.)* for DNA or RNA sequencing.

1. **Related Documents**

| **Title** | **Document Control Number** |
| --- | --- |
| MiSeq Preventive Maintenance Wash Flowchart |  |
| MiSeq In-Use EquipmentMaintenance Log |  |
| MiSeq Standby Equipment Maintenance Log |  |
| Equipment Out of Service Form |  |
| Master Equipment Inventory Log |  |
| Master Maintenance / Calibration Schedule |  |

1. **Responsibilities**

|  |  |
| --- | --- |
| **Position** | **Responsibility** |
| All laboratory staff | * Ensures equipment is properly maintained according to established criteria
* Follows documented equipment procedures
 |
| Branch Chief / Team Lead | * Ensures documented procedures for the proper maintenance of designated equipment are established
* Ensures documented procedures are followed
 |
| Quality Manager | * Ensures documented equipment procedures are available to the end user
* Maintains a master list of equipment used by the laboratory
 |

1. **Definitions**

|  |  |
| --- | --- |
| **Term** | **Definition** |
| Preventive maintenance | [Systematic](http://www.businessdictionary.com/definition/systematic.html) [inspection](http://www.businessdictionary.com/definition/inspection.html), detection, [correction](http://www.businessdictionary.com/definition/correction.html), and [prevention](http://www.businessdictionary.com/definition/prevention.html) of incipient [failures](http://www.businessdictionary.com/definition/failure.html) for the purpose of preventing actual or major failures. |

1. **Equipment / Materials**

|  |  |  |
| --- | --- | --- |
| **Supply** | **Catalog Number** | **Procedure** |
| Tween 20 | Sigma-Aldrich Cat. # P7949 or equivalent | * Post-Run Wash
* Maintenance Wash
* Standby Wash
 |
| Laboratory Grade Water | N/A | * Post-Run Wash
* Maintenance Wash
* Standby Wash
 |
| 6% Sodium hypochlorite | N/A | * Post-Run Wash with Template Line Wash
 |
| MiSeq Tube | Illumina Part # MS-102-9999 | * Post-Run Wash with Template Line Wash
 |

1. **Safety Precautions**
	* 1. The MiSeq waste bottle contains formamide, an aliphatic amide that is a probable reproductive toxin. Personal injury can occur through inhalation, ingestion, skin contact, and eye contact.
2. **Procedure**
	1. **Maintenance**
		1. Any update to the equipment, inclusive of software updates, requires evaluation and approval prior to installation. Performance of Installation, Operational, and possibly Performance Qualification may be required.
		2. Reference the *MiSeq Preventive Maintenance Wash Flowchart* for additional guidance on maintenance wash requirements.
	2. **Weekly Maintenance**
		1. **Cleaning**
			1. Cleaning should be performed weekly.
			2. Using a Kimwipe®, wipe the outer casing to remove dust. Do not touch the instrument if it is running.
			3. Record in *laboratory cleaning / maintenance log*.
		2. **Power Cycle the Instrument**
			1. To power cycle in Windows mode:
				* Close MiSeq Control Software and ensure no other programs are running and shut-down the computer from the Windows Start button.
				* Once the computer has shut down, turn off the power switch on the back of the MiSeq instrument, and leave it off for a minimum of 60 seconds.
				* Turn on the power switch and let the computer start normally.
				* Record in *laboratory cleaning / maintenance log*.
			2. To power cycle in Kiosk mode:
				* Go to **Manage Instrument** andselect **Shutdown**
				* Once the computer has shut down, turn off the power switch on the back of the MiSeq instrument and leave it off for a minimum of 60 seconds.
				* Turn on the power switch and let the computer start normally.
				* Record in *laboratory cleaning / maintenance log*.
	3. **Post-Run Wash**
		1. A post-run wash must be completed after each sequencing run. The post-run wash takes approximately 20 minutes to complete.
		2. Leave the used flow cell on the instrument.
		3. Prepare fresh wash solution with Tween 20 and laboratory-grade water.
			1. Add 10 ml 100% Tween 20 to 90 ml laboratory grade water. These volumes result in 10% Tween 20.
			2. Add 50 ml 10% Tween 20 to 950 ml laboratory grade water. These volumes result in a 0.5% Tween 20 wash solution.
			3. Invert five times to mix.
		4. Prepare the wash components with fresh wash solution, as follows:
			1. Add wash solution to each reservoir of the wash tray, fill each reservoir to 90% capacity or 6 ml of wash solution into each reservoir.
			2. Add 350 ml wash solution to the 500 ml wash bottle.
		5. When the sequencing run is complete, select **Start Wash**. The software automatically raises the sippers in the reagent chiller.
		6. Open the reagent compartment door and reagent chiller door, wait for sippers to raise and slide the used reagent cartridge from the chiller.
		7. Slide the wash tray into the reagent chiller until it stops, then close the reagent chiller door.
		8. Raise the sipper handle in front of the PR2 bottle and waste bottle until it locks into place.
		9. Remove the PR2 bottle and replace it with the wash bottle.
			1. **Discard the PR2 bottle in a laboratory hazardous waste container.** Do not reuse any remaining PR2.
		10. Remove the waste bottle and discard the contents appropriately. **(Caution: waste contains formamide; discard using the CDC Hazardous waste turn-in system.)** Return the waste bottle to the reagent compartment.
		11. Slowly lower the sipper handle, making sure that the sippers lower into the wash bottle and waste bottle, proceed to close the reagent compartment door.
		12. Select **Next**. The post-run wash begins.
		13. When the wash is complete, leave the used flow cell, wash tray, and wash bottle containing the remaining wash solution on the instrument.
		14. Record the following on the *MiSeq In-Use Equipment Maintenance Log*:
			1. Sequence run start date
			2. Post-run wash date
			3. Indication that post-run wash did not include template line wash (N)
			4. Initials of operator performing maintenance
	4. **Post-Run Wash with Template Line Wash**
		1. MCS v2.5 or higher is required to perform this post-run wash with template line wash procedure. A post-run wash with template line wash is completed after each sequencing run. The post-run wash with template line wash takes approximately 30 minutes to complete.
		2. Leave the used flow cell on the instrument.
		3. Prepare fresh wash solution with Tween 20 and laboratory-grade water.
			1. Add 10 ml 100% Tween 20 to 90 ml laboratory grade water. These volumes result in 10% Tween 20.
			2. Add 50 ml 10% Tween 20 to 950 ml laboratory grade water. These volumes result in a 0.5% Tween 20 wash solution.
			3. Invert five times to mix.
		4. Prepare fresh sodium hypochlorite wash solution with laboratory grade water, as follows:
			1. Add 36 µl of 5% sodium hypochlorite to 864 µl laboratory grade water. These volumes result in a 1:25 sodium hypochlorite dilution.
			2. Add 50 µl of the 1:25 sodium hypochlorite dilution to 950 µl of laboratory grade water in an Illumina supplied MiSeq tube.
		5. Prepare the wash components with fresh wash solution, as follows:
			1. Add wash solution to each reservoir of the wash tray, fill each reservoir to 90% capacity or 6 ml of wash solution into each reservoir.
			2. Add 350 ml wash solution to the 500 ml wash bottle.
		6. Insert the MiSeq tube containing 0.01% sodium hypochlorite wash solution into position 17 of the wash tray until the neck of the tube is flush with the tray.
		7. When the sequencing run is complete, select **Start Wash**. The software automatically raises the sippers in the reagent chiller.
		8. Select **Perform optional template line wash** on the Post-Run Wash screen.
		9. Open the reagent compartment door and reagent chiller door, wait for sippers to raise, and slide the used reagent cartridge from the chiller.
			1. Slide the wash tray into the reagent chiller until it stops, and then close the reagent chiller door.
		10. Raise the sipper handle in front of the PR2 bottle and waste bottle until it locks into place.
		11. Remove the PR2 bottle and replace it with the wash bottle.
			1. **Discard the PR2 bottle in a laboratory hazardous waste container.** Do not reuse any remaining PR2
		12. Remove the waste bottle and discard the contents appropriately. **(Caution: waste contains formamide; discard using the CDC Hazardous waste turn-in system.)** Return the waste bottle to the reagent compartment.
		13. Slowly lower the sipper handle, making sure that the sippers lower into the wash bottle and waste bottle and proceed to close the reagent compartment door.
		14. Select **Next**. The post-run wash with template line wash begins.
		15. When the wash is complete, leave the used flow cell, wash tray, and wash bottle containing the remaining wash solution on the instrument.
		16. Record the following on the *MiSeq In-Use Equipment Maintenance Log*:
			1. Sequence run start date
			2. Post-run wash date
			3. Indication that the post-run wash did include template line wash (Y)
			4. Initials of operator performing maintenance
	5. **Maintenance Wash**
		1. A maintenance wash must be completed every 30 days. Additionally, a maintenance wash must be completed if the instrument has been in standby mode. The maintenance wash takes approximately 90 minutes to complete.
		2. Leave the used flow cell on the instrument.
		3. From the Home screen, select **Perform Wash**.
		4. From the Perform Wash screen, select **Maintenance Wash**. The software automatically raises the sippers in the reagent chiller.
		5. **Perform First Wash**
			1. Prepare fresh wash solution with Tween 20 and laboratory grade water as follows:
				* Add 10 ml 100% Tween 20 to 90 ml laboratory grade water. These volumes result in 10% Tween 20.
				* Add 50 ml 10% Tween 20 to 950 ml laboratory grade water. These volumes result in a 0.5% Tween 20 wash solution.
				* Invert five times to mix.
			2. Prepare the wash components with fresh wash solution as follows:
				* Add wash solution to each reservoir of the wash tray, fill each reservoir to 90% capacity or 6 ml of wash solution into each reservoir.
				* Add 350 ml wash solution to the 500 ml wash bottle.
			3. Open the reagent compartment door and reagent chiller door, wait for sippers to raise, and slide the used reagent cartridge from the chiller.
			4. Slide the wash tray into the reagent chiller until it stops, and then close the reagent chiller door.
				* Raise the sipper handle in front of the PR2 bottle and waste bottle until it locks into place.
			5. Remove the PR2 bottle and replace it with the wash bottle.
				* **Discard the PR2 bottle in a laboratory hazardous waste container.** Do not reuse any remaining PR2
			6. Remove the waste bottle and discard the contents appropriately. **(Caution: waste contains formamide; discard using the CDC Hazardous waste turn-in system.)** Return the waste bottle to the reagent compartment.
			7. Slowly lower the sipper handle, making sure that the sippers lower into the wash bottle and waste bottle. Close the reagent compartment door and Select **Next**. The first wash begins.
		6. **Perform Second Wash**
			1. **Always use fresh wash solution in the wash tray for each wash step.**
			2. When the first wash is complete, remove the wash tray and discard the remaining wash solution.
			3. Refill the wash components with fresh wash solution, as follows:
				* Add wash solution to each reservoir of the wash tray, fill each reservoir to 90% capacity or 6 ml of wash solution into each reservoir.
			4. Slide the wash tray into the reagent chiller until it stops, and then close the reagent chiller door.
			5. Close the reagent compartment door.
			6. Select **Next**. The second wash begins.
		7. **Perform Final Wash**
			1. **Always use fresh wash solution in the wash tray for each wash step.**
			2. When the second wash is complete, remove the wash tray and discard the remaining wash solution.
			3. Refill the wash components with fresh wash solution, as follows:
				* Add wash solution to each reservoir of the wash tray, fill each reservoir to 90% capacity or 6 ml of wash solution into each reservoir.
			4. Slide the wash tray into the reagent chiller until it stops, and then close the reagent chiller door.
			5. Close the reagent compartment door.
			6. Select **Next**. The final wash begins.
			7. When the wash is complete, leave the used flow cell, wash tray, and wash bottle containing the remaining wash solution on the instrument.
			8. Record the following on the *MiSeq In-Use Equipment Maintenance Log:*
				* Maintenance wash date
				* Initials of the operator performing maintenance
	6. **Standby Wash**
		1. A standby wash must be completed if there are no plans to use the instrument within the next seven days. Additionally, a standby wash must be completed every 30 days until the instrument is brought back into use through a maintenance wash. The standby wash takes approximately 2 hours to complete.
		2. Leave the used flow cell on the instrument.
		3. From the Home screen, select **Perform Wash**.
		4. From the Wash Options screen, select **Standby Wash**. The software automatically raises the sippers in the reagent chiller.
		5. **Perform First Wash**
			1. Prepare fresh wash solution with Tween 20 and laboratory-grade water.
				* Add 10 ml 100% Tween 20 to 90 ml laboratory grade water. These volumes result in 10% Tween 20.
				* Add 50 ml 10% Tween 20 to 950 ml laboratory grade water. These volumes result in a 0.5% Tween 20 wash solution.
				* Invert five times to mix.
			2. Prepare the wash components with fresh wash solution, as follows:
				* Add wash solution to each reservoir of the wash tray, fill each reservoir to 90% capacity or 6 ml of wash solution into each reservoir.
				* Add 350 ml wash solution to the 500 ml wash bottle.
			3. Open the reagent compartment door and reagent chiller door, wait for sippers to raise, and slide the used reagent cartridge from the chiller.
			4. Slide the wash tray into the reagent chiller until it stops, and then close the reagent chiller door.
				* Raise the sipper handle in front of the PR2 bottle and waste bottle until it locks into place, and replace the PR2 bottle with the wash bottle.
			5. **Discard the PR2 bottle in a laboratory hazardous waste container.** Do not reuse any remaining PR2
			6. Remove the waste bottle and discard the contents appropriately. **(Caution: waste contains formamide; discard using the CDC Hazardous waste turn-in system.)** Return the waste bottle to the reagent compartment.
			7. Slowly lower the sipper handle, making sure that the sippers lower into the wash bottle and waste bottle.
			8. Close the reagent compartment door.
			9. Select **Next**. The first wash begins.
		6. **Perform Second Wash**
			1. **Always use fresh wash solution in the wash tray for each wash step.**
			2. When the first wash is complete, remove the wash tray and discard the remaining wash solution.
			3. Refill the wash components with fresh wash solution, as follows:
				* Add wash solution to each reservoir of the wash tray, fill each reservoir to 90% capacity or 6 ml of wash solution into each reservoir.
			4. Slide the wash tray into the reagent chiller until it stops, and then close the reagent chiller door.
			5. Close the reagent compartment door.
			6. Select **Next**. The second wash begins.
			7. When the wash is complete, leave the used flow cell, wash tray, and wash bottle containing the remaining wash solution on the instrument.
			8. Record the following on the *MiSeq Standby Equipment Maintenance Log:*
				* Standby wash date
				* Initials of operator performing maintenance
	7. **Repair / Service / Unscheduled Maintenance**

*NOTE: if your laboratory has an equipment troubleshooting or Out of Service SOP, delete the text below, include a reference to the SOP, and add the SOP as a related document in Section 3.0.*

* + - 1. Place an “Out of Service (OOS)” form on the equipment.
			2. Document the problem on the *laboratory OOS / maintenance log*, stating date / time taken OOS, reason why the equipment was taken OOS, and initials / date of responsible individual.
			3. “Troubleshoot” source of the problem (sample, reagent, operator, equipment, etc.). (Refer to Illumina MiSeq System User Guide.)
			4. Call Manufacturer’s Technical Assistance, if needed. Record the technical support case number.
			5. Determine what repair / maintenance is to be performed when you call for service.
				* Equipment under warranty may require that repairs are completed by the manufacturer.
				* Is disinfection / decontamination required?
				* How is disinfection / decontamination performed? Define appropriate disinfectant, time required, recommended precautions, areas to be decontaminated, etc.
			6. Items sent to a manufacturer for repair and ultimately replaced must be reported to the responsible property office.
			7. Record in *laboratory OOS / maintenance log* and attach service report, if applicable.
			8. Place equipment back into service after verification / qualification completed.
			9. Maintain a history of maintenance / repair / service.

1. **References**
	1. CLSI, Laboratory Implementation, Verification & Maintenance: Approved Guideline GP31-A.
	2. Illumina MiSeq System Guide Document #15027617 v04 July 2018
2. **Revision History**

| **Rev #** | **DCR #** | **Change Summary** | **Date** |
| --- | --- | --- | --- |
|  |  |  |  |

1. **Approval**

Reviewed By: \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_ Date: \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_