1. Sample preparation for MagMAX™CORE

- Note: Throughout this document, 1x PBS pH=7.4 is referred to simply as PBS

1 Before start

Before starting the protocol, make sure that you have 70% ethanol, your sample map and all materials needed.

1.1 Turn on the safe cabinet (motor and white light).
1.2 Clean the safe cabinet (work surface, walls and glass) using 70% ethanol.
1.3 Turn off the white light and turn on the UV light for 15 minutes.
1.4 Turn of the UV light and start working.
1.5 Clean all materials with 70% ethanol before putting them into the cabinet.

2. Materials

2.1 Sterile centrifuge tubes
2.2 PBS pH=7.4
2.3 Single Channel Pipette (1000 μL)
2.4 Pipette Tips (1000 μL)
2.5 Tweezers
2.6 Becker with 70% Ethanol
2.7 Sharper marker

3. Sample Preparation

3.1 Add 01 mL of PBS to each tube containing the swab.
3.2 Homogenize tubes for 30 minutes.
3.3 Transfer swabs to new tubes. Keep them frozen.
3.4 Centrifuge tubes containing the homogenized samples at 14 000 X g for 30 minutes.
3.5 Suspend pellets in 300 □l of PBS.
3.6 Homogenize tubes for 30 minutes.
3.7 Keep samples at -80ºC until processing.
3.8 **Use the suspensions in the MagMAX™ CORE extraction protocols**

2. Extraction of WGS Bacterial Isolates Using MagMAX™ CORE Mechanical Lysis Module and MagMAX™ CORE (Kit 1)

1. Purpose

The purpose of this procedure is to define a uniform method of extraction of DNA and RNA from different sample type using MagMAX™ CORE Nucleic acid purification kit (cat. #A32700 or A32702).

2. Scope

This procedure is applicable fecal samples requiring DNA and RNA extraction. This procedure utilizes the MagMAX™ CORE Nucleic Acid Purification Kit (Thermo, cat. #A32700 or A32702).

3. Before start

Before starting the protocol, make sure that you have 70% ethanol, your sample map and all materials needed.

3.1 Turn on the safe cabinet (motor and white light).
3.2 Clean the safe cabinet (work surface, walls and glass) using 70% ethanol.
3.3 Turn off the white light and turn on the UV light for 15 minutes.
3.4 Turn of the UV light and start working.
3.5 Clean all materials with 70% ethanol before putting them into the cabinet.

4. Materials

Separate the following materials in a cart:

4.1 MagMAX™ CORE Nucleic Acid Purification Kit
4.2 MagMAX™ CORE Mechanical Lysis Module Kit
4.3 Single Channel Pipettes (2.5 μL, 10 μL, 100 μL, 200 μL and 1000 μL)
4.4 Multi-Channel Pipettes (100 μL, 300 μL)
4.5 Pipette Tips (10 μL, 100 μL, 200 μL, 300 μL and 1000 μL)
4.6 50 mL or 15 mL tubes (depending on the number of samples)
4.7 Graduated pipette (depending on the number of samples)
4.8 Tube rack
4.9 Reagent Reservoir
4.10 03 Deep well plates (Wash 1, Wash 2 and Sample)
4.11 01 Standard plate (Elution)
4.12 01 Tip Comb
4.13 Plate film
4.14 Trash bag
4.15 Sharp marker

4. Plate set up

Plate ID	Plate Type	Reagent	Volume per well
Tip Comb	Standard plate	Place a tip comb in the plate.	
Sample plate	Deep well	Sample, CORE Lysis/binding Solution, ProteinaseK	
Wash 1	Deep well	MagMAX™ CORE Wash Solution 1	**500 µl**
Wash 2	Deep well	MagMAX™ CORE Wash Solution 2	**500 µl**
Elution (label with worklist date)	Standard	MagMAX™ CORE Elution Buffer	**90 µl**

Deep well plates (ThermoFisher Scientific, cat. #10045), standard plates (Thermo, cat. # 97002540), and deep-well plate comb (Thermo, cat. # 97002534)

5. Reagent and plate preparation

5.1 All components of this kit are stored at room temperature.

5.2 Make Lysis/Biding/Beads mix solution as per MagMAX™ CORE Lysis Solution, MagMAX™ CORE Biding Solution and MagMAX™ CORE Magnetic Beads, which includes 350 µl/reaction of MagMAX™ CORE Lysis Solution, 350 µl/reaction of MagMAX™ Core Binding Solution, and 20 µl/reaction of MagMAX™ CORE Magnetic Beads. After the addition of each component of the Lysis/Biding/Beads mix solution, be sure to thoroughly vortex the solution. Keep the solution at room temperature. The prepared lysis buffer can be stored at room temperature for up to 24 hours.

5.3 Prepare Wash 1 plate by adding 500 µL of MagMAX™ CORE Wash Solution 1 to each well of a deep well plate (make sure that you label the plate as WASH 1).

5.4 Prepare Wash 2 plate by adding 500 µL of MagMAX™ CORE Wash Solution 2 to each well of a deep well plate (make sure that you label the plate as WASH 2).

5.5 Prepare Elution plate by adding 90 µL of MagMAX™ CORE Elution Buffer to each well of a standard plate (make sure that you label the plate with worklist date).

6. Sample preparation

6.1 Add 400 µl of **MagMAX™ CORE Clarifying Solution** to the required number of MagMAX™ CORE Bead Beating Tubes.

6.2 Add 175 µl of concentrated sample to the MagMAX™ CORE Bead Beating Tube.

6.3 Disrupt (bead-beating) the samples (For Bead Ruptor 96, homogenize samples for 05 minutes at 20 Hz).

6.4 Centrifuge MagMAX™ CORE Bead Beating Tube at 15,000 x g for 3 minutes.

6.5 In the Sample plate:

6.5.1 Add 10 µl of **MagMAX™ CORE Proteinase K** to each well of the plate.

6.5.2 Add 300 µl of supernatant (clarified lysate).

6.5.3 Mix the Clarified lysate with proteinase K by pipetting up and down several times.

6.5.4 Incubate for 2 minutes at room temperature.

6.5.5 Add 720 µl of Lysis/Binding/Beads Mix to each sample. Immediately proceed to magnetic particle processor (KingFisher Flex).

7. Setting up the equipment

7.1 Turn on the KingFisher Flex. Push the right arrow to select “user” then the down arrow to select “DNA”. Toggle down to the “MagMAX_Core_Flex” program and then push start.

7.2 Load each of the plates according to the display’s instructions, pressing start after each plate is loaded. Be sure the orientation of the plate is correct (A1 is in the A1 location).

7.3 Press start and record usage in the logbook.

7.4 The program runs approximately 27 minutes. The first plate to be removed is the Elution plate, which now contains the eluted DNA. Place a lid over this plate, label it with the worklist date, and seal it with parafilm. Place the plate with the eluted DNA on ice. All other plates can be placed in a ziplock bag and discarded in the biohazard waste, except for the unlabeled plate that held the comb, which can be reused.

7.5 Turn off the KingFisher Flex and wipe down with DNA away solution (Thermo Scientific, cat. # 7000).

7.6 The elution plate can now be used for bacterial whole genome sequencing methods.

8. References

Applied Biosystems, Instruction Manual for MagMAX™ CORE Nucleic acid purification kit (cat. #A32700 or A32702)

3. Extraction of WGS Bacterial Isolates Using MagMAX™ Microbiome Ultra Nucleic Acid Isolation Kit (kit 2)

1. Purpose

The purpose of this procedure is to define a uniform method of extraction of DNA and RNA from fecal samples using the MagMAX™ Microbiome Ultra Nucleic Acid Isolation kit (cat. # A42358).

2. Scope

This procedure is applicable to fecal samples requiring DNA and RNA extraction. This procedure utilizes the MagMAX™ Microbiome Ultra Nucleic Acid Isolation Kit (Thermo, cat. # A42358).

3. Before start

3.1 Turn on the safe cabinet (motor and white light).
3.2 Clean the safe cabinet (work surface, walls and glass) using 70% ethanol.
3.3 Turn off the white light and turn on the UV light for 15 minutes.
3.4 Turn of the UV light and start working.
3.5 Clean all materials with 70% ethanol before putting them into the cabinet.

4. Materials

Separate the following materials in a cart:

4.1 MagMAX™ CORE Microbiome Ultra Nucleic Acid Purification Kit
4.2 100% absolute Ethanol
4.3 Nuclease-Free Water
4.4 Single Channel Pipettes (2.5 μL, 10 μL, 100 μL, 200 μL and 1000 μL)
4.5 Multi-Channel Pipettes (100 μL, 300 μL)
4.6 Pipette Tips (10 μL, 100 μL, 200 μL, 300 μL and 1000 μL)
4.7 50 mL or 15 mL tubes (depending on the number of samples)
4.8 Graduated pipette (depending on the number of samples)
4.9 Tube rack
4.10 Reagent Reservoir
4.11 06 Deep well plates (Wash 1, Wash 2, Wash 3, Wash 4 and Sample)
4.12 01 Tip Comb
4.13 Plate film
4.14 Trash bag
4.15 Sharp marker

5. Plate set up

Plate ID	Plate Type	Reagent	Volume per well
Tip Comb	Standard plate	Place a 96 Deep-well tip comb in a Standard deep-well plate.	
Sample plate	Deep well	Sample, Biding Bead Mix	
Wash 1	Deep well	Wash Buffer	**1000 µl**
Wash 2	Deep well	Wash Buffer	**1000 µl**
Wash 3	Deep well	80% Ethanol	**1000 µl**
Wash 4	Deep well	80% Ethanol	**1000 µl**
Elution (label with worklist date)	Deep well	Elution Solution	**200**

Deep well plates (ThermoFisher Scientific, cat. #10045), standard plates (Thermo, cat. # 97002540), and deep-well plate comb (Thermo, cat. # 97002534)

6. Reagent and plate preparation

6.1 All components of this kit are stored at room temperature.

6.2 Before first use of the kit:

6.2.1 Prepare 80% Ethanol from 100% absolute Ethanol and Nuclease-Free Water prepare enough solution for a minimum volume of 2 mL per sample.

6.3 Before each use of the kit:

6.3.1 Make Binding Bead mix solution as per Viral/Pathogen Biding Solution and DNA/RNA Biding Beads recipe, which includes 500 µl/reaction Viral/Pathogen Biding Solution and 20 µl/reaction of DNA/RNA Biding Bead.

6.3.2 After the addition of each component of the Binding/Bead Solution, be sure to thoroughly vortex the solution. Keep the solution at room temperature.

6.4 Prepare Wash 1 plate by adding 1000 µL of MagMAX™ MICROBIOME Wash Buffer to each well of a deep well plate (make sure that you label the plate as WASH 1).

6.5 Prepare Wash 2 plate by adding 1000 µL of MagMAX™ MICROBIOME Wash Buffer to each well of a deep well plate (make sure that you label the plate as WASH 2).

6.6 Prepare Wash 3 plate by adding 1000 µL of 80% Ethanol to each well of a deep well plate (make sure that you label the plate as WASH 3).

6.7 Prepare Wash 4 plate by adding 1000 µL of 80% Ethanol to each well of a deep well plate (make sure that you label the plate as WASH 4)

6.8 Prepare Elution plate by adding 200 µL of MagMAX™ MICROBIOME Elution Solution to each well of a standard plate (make sure that you label the plate with worklist date).

7. Sample preparation

7.1 Set up the Vortex (Scientific Industries # SI-0236) with the vortex adapter (Scientific Industries # SI-H524).

7.2 Add 800 µl of Lysis Buffer to the bead tubes.

7.3 Remove the plastic stick. Then, place the swab into the prepared bead tube.

7.4 Cap, then vortex the bead tube upside down for 10 seconds to mix the sample with the buffer.

7.5 Set the Vortex speed to 2,500 rpm. Then, place the tubes onto the adapter.

7.6 Lyse samples on the Vortex for 10 minutes (Bead tube can be stored at 4ºC overnight after lysis).

7.7 Remove tubes from the Vortex and centrifuge for 2 minutes at 13,200 x *g*.

7.8 Transfer 400 µl of the sample to the appropriate wells of the Sample Plate (Lysate can be stored at -20ºC for up to 3 months. Before use, ensure sample has equilibrated to room temperature).

7.9 Invert Biding Bead Mix to mix. Then, add 520 µl to each sample in the Sample Plate. Immediately proceed to magnetic particle processor (KingFisher Flex).

8. Setting up the equipment

8.1 Turn on the KingFisher Flex. Push the right arrow to select “user” then the down arrow to select “DNA”. Toggle down to the “MagMAX_Microbiome_Stool_Flex” program and then push start.

8.2 Load each of the plates according to the display’s instructions, pressing start after each plate is loaded. Be sure that the orientation of the plate is correct (A1 is in the A1 location).

8.3 Press start and record usage in the logbook

8.4 The program runs approximately 35 minutes. The first plate to be removed is the Elution plate, which now contains the eluted Nucleic Acid. Place a lid over this plate, label it with the worklist date, and seal it with parafilm. Place the plate with the eluted Nucleic Acid on ice. All other plates can be placed in a ziplock bag and discarded in the biohazard waste, with the exception of the unlabeled plate that held the comb, which can be reused.

8.5 Turn off the KingFisher Flex and wipe down with DNA Away solution (Thermo Scientific, cat. # 7000).

8.6 The elution plate can now be used for bacterial whole genome sequencing methods.

9. References

Applied Biosystems, Instruction Manual for MagMAX™ Microbiome Ultra Nucleic Acid Isolation Kit (cat. #A42358)

4. Extraction of WGS Bacterial Isolates Using In-house Bead Loading System and MagMAX™ CORE with Clarifying Solution (kit 3)

1.0 Purpose

The purpose of this procedure is to define a uniform method of extraction of DNA and RNA from different sample type using an in -house bead loading system and MagMAX™ CORE Nucleic acid purification kit (cat. #A32700 or A32702).

2.0 Scope

This procedure is applicable to fecal samples requiring DNA and RNA extraction. This procedure utilizes the MagMAX™ CORE Nucleic Acid Purification Kit (Thermo, cat. #A32700 or A32702).

3. Before start

Before starting the protocol, make sure that you have 70% ethanol, your sample map and all materials needed.

3.1 Turn on the safe cabinet (motor and white light).
3.2 Clean the safe cabinet (work surface, walls and glass) using 70% ethanol.
3.3 Turn off the white light and turn on the UV light for 15 minutes.
3.4 Turn of the UV light and start working.
3.5 Clean all materials with 70% ethanol before putting them into the cabinet.

4. Materials

Separate the following materials in a cart:

4.1 MagMAX™ CORE Nucleic Acid Purification Kit
4.2 Single Channel Pipettes (2.5 μL, 10 μL, 100 μL, 200 μL and 1000 μL)
4.3 Multi-Channel Pipettes (100 μL, 300 μL)
4.4 Pipette Tips (10 μL, 100 μL, 200 μL, 300 μL and 1000 μL)
4.5 50 mL or 15 mL tubes (depending on the number of samples)
4.6 Graduated pipette (depending on the number of samples)
4.7 Tube rack
4.8 Reagent Reservoir
4.9 03 Deep well plates (Wash 1, Wash 2 and Sample)
4.10 01 Standard plate (Elution)
4.11 01 Tip Comb
4.12 Plate film
4.13 Zirconia tubes
4.14 Decapper
4.15 Trash bag
4.16 Sharp marker

5. Plate set up

Plate ID	Plate Type	Reagent	Volume per well
Tip Comb	Standard plate	Place a tip comb in the plate. Be sure the comb is flat	
Sample plate	Deep well	Bead mix, lysate and CORE binding Solution	
Wash 1	Deep well	MagMAX™ CORE Wash Solution 1	**500 µl**
Wash 2	Deep well	MagMAX™ CORE Wash Solution 2	**500 µl**
Elution (label with worklist date)	Standard	MagMAX™ CORE Elution Buffer	**90 µl**

Deep well plates (ThermoFisher Scientific, cat. #10045), standard plates (Thermo, cat. # 97002540), and deep-well plate comb (Thermo, cat. # 97002534)

6. Reagent and plate preparation

6.1 All components of this kit are stored at room temperature.

6.2 Make Lysis solution as per MagMAX™ CORE Lysis Solution, which includes 450 µl/reaction of MagMAX™ CORE Lysis Solution and 2.05 µl/reaction of Clarifying Solution (Qiagen, cat. # 19088). After the addition of each component of the Lysis Buffer, be sure to thoroughly vortex the solution. Keep the solution at room temperature. The prepared lysis buffer can be stored at room temperature for up to 24 hours.

6.3 Prepare Bead/Proteinase K Mix as per recipe: 20 µl/reaction of MagMAX™ CORE Magnetic Beads and 10 µl/reaction of MagMAX™ CORE Proteinase K (20 mg/mL). It's recommended to prepare Bead/Proteinase K mix daily, but this can be stored at 4°C for up to one week.

6.4 Prepare Wash 1 plate by adding 500 µL of MagMAX™ CORE Wash Solution 1 to each well of a deep well plate (make sure that you label the plate as WASH 1).

6.5 Prepare Wash 2 plate by adding 500 µL of MagMAX™ CORE Wash Solution 2 to each well of a deep well plate (make sure that you label the plate as WASH 2).

6.6 Prepare Elution plate by adding 90 µL of MagMAX™ CORE Elution Buffer to each well of a standard plate (make sure that you label the plate with worklist date).

7. Sample preparation

7.1 Add 450 µl of lysis buffer (step 5.2) to each zirconia bead tube corresponding to a sample.

7.2 Pipet to mix and transfer 200 µl of sample to the zirconia tube in a biological safety cabinet.

7.3 Be sure that all caps are inserted into the tubes to the same depth.

7.4 Place the bead tube rack in the Bead-Ruptor 96 (Omni, cat. #SKU: 27- 0001) using the appropriate lid.

7.5 Bead beat the samples at 20 Hz for 2.5 min. Rest for 5 min, and bead beat for an additional at 20 Hz for 2.5 min. Be sure to switch off and unplug the Bead-Ruptor 96.

7.6 Centrifuge samples at 2,500 rpm for 5 min (Be sure to put the plate rotor correctly).

7.7 Invert the tube of Bead/Proteinase K and vortex to mix several times to resuspend the beads. Then add 30 µl of Bead/Proteinase K mix to the required wells on the deep well plate labelled "Sample".

7.8 Using the decapper (ISC BioExpress, cat. # P-8754-04), remove the caps from the tubes. Discard the caps into the waste container.

7.9 Transfer 500 µl of lysate from bead tubes to the sample plate with bead mix. Be careful not to transfer any zirconia/silica beads. Seal the plate

7.10 Using the ThermoMixer C (Eppendorf, cat. # 2231000574) shake sealed sample plate at 1400 rpm for 2 min.

7.11 Unseal the sample plate and add 350 µl of **MagMAX™ CORE Binding Solution** to each sample well. Immediately proceed to magnetic particle processor (KingFisher Flex).

8. Setting up the equipment

8.1 Turn on the KingFisher Flex. Push the right arrow to select "user" then the down arrow to select "DNA". Toggle down to the "MagMAX_Core_Flex" program and then push start.

8.2 Load each of the plates according to the display's instructions, pressing start after each plate is loaded. Be sure the orientation of the plate is correct (A1 is in the A1 location).

8.3 Press start and record usage in the logbook.

8.4 The program runs approximately 27 minutes. The first plate to be removed is the Elution plate, which now contains the eluted DNA. Place a lid over this plate, label it with the worklist date, and seal it with parafilm. Place the plate with

the eluted DNA on ice. A**l** other plates can be placed in a ziplock bag and discarded in the

biohazard waste, except for the unlabeled plate that held the comb, which can be reused.

8.5 Turn off the KingFisher Flex and wipe down with DNA away solution (Thermo Scientific, cat. # 7000).

8.6 **The elution plate can now be used for bacterial whole genome sequencing methods.**

9. References

Applied Biosystems, Instruction Manual for MagMAX™ CORE Nucleic acid purification kit (cat. #A32700 or A32702)
AAVLD Section 5.4 Test
Methods ISO 17025
Section 5.4 Test
Methods
AHDC QM Section 5.4 Test Methods and Method Validation

5. Extraction of WGS Bacterial Isolates Using In-house Bead Loading System and MagMAX™ CORE with Reagent DX (kit 4)

1.0 Purpose

The purpose of this procedure is to define a uniform method of extraction of DNA and RNA from different sample type using an in -house bead loading system and MagMAX™ CORE Nucleic acid purification kit (cat. #A32700 or A32702).

2.0 Scope

This procedure is applicable to fecal samples requiring DNA and RNA extraction. This procedure utilizes the MagMAX™ CORE Nucleic Acid Purification Kit (Thermo, cat. #A32700 or A32702).

3. Before start

Before starting the protocol, make sure that you have 70% ethanol, your sample map and all materials needed.

3.6 Turn on the safe cabinet (motor and white light).
3.7 Clean the safe cabinet (work surface, walls and glass) using 70% ethanol.
3.8 Turn off the white light and turn on the UV light for 15 minutes.
3.9 Turn of the UV light and start working.
3.10 Clean all materials with 70% ethanol before putting them into the cabinet.

4. Materials

Separate the following materials in a cart:

4.17 MagMAX™ CORE Nucleic Acid Purification Kit
4.18 Single Channel Pipettes (2.5 μL, 10 μL, 100 μL, 200 μL and 1000 μL)
4.19 Multi-Channel Pipettes (100 μL, 300 μL)
4.20 Pipette Tips (10 μL, 100 μL, 200 μL, 300 μL and 1000 μL)
4.21 50 mL or 15 mL tubes (depending on the number of samples)
4.22 Graduated pipette (depending on the number of samples)
4.23 Tube rack
4.24 Reagent Reservoir
4.25 03 Deep well plates (Wash 1, Wash 2 and Sample)
4.26 01 Standard plate (Elution)
4.27 01 Tip Comb
4.28 Plate film
4.29 Zirconia tubes
4.30 Decapper
4.31 Trash bag
4.32 Sharp marker

10. Plate set up

Plate ID	Plate Type	Reagent	Volume per well
Tip Comb	Standard plate	Place a tip comb in the plate. Be sure the comb is flat	
Sample plate	Deep well	Bead mix, lysate and CORE binding Solution	
Wash 1	Deep well	MagMAX™ CORE Wash Solution 1	**500 µl**
Wash 2	Deep well	MagMAX™ CORE Wash Solution 2	**500 µl**
Elution (label with worklist date)	Standard	MagMAX™ CORE Elution Buffer	**90 µl**

Deep well plates (ThermoFisher Scientific, cat. #10045), standard plates (Thermo, cat. # 97002540), and deep-well plate comb (Thermo, cat. # 97002534)

11. Reagent and plate preparation

11.1 All components of this kit are stored at room temperature.

11.2 Make Lysis solution as per MagMAX™ CORE Lysis Solution, which includes 450 µl/reaction of MagMAX™ CORE Lysis Solution and 2.05 µl/reaction of Reagent DX (Qiagen, cat. # 19088). After the addition of each component of the Lysis Buffer, be sure to thoroughly vortex the solution. Keep the solution at room temperature. The prepared lysis buffer can be stored at room temperature for up to 24 hours.

11.3 Prepare Bead/Proteinase K Mix as per recipe: 20 µl/reaction of MagMAX™ CORE Magnetic Beads and 10 µl/reaction of MagMAX™ CORE Proteinase K (20 mg/mL). It's recommended to prepare Bead/Proteinase K mix daily, but this can be stored at 4°C for up to one week.

11.4 Prepare Wash 1 plate by adding 500 µL of MagMAX™ CORE Wash Solution 1 to each well of a deep well plate (make sure that you label the plate as WASH 1).

11.5 Prepare Wash 2 plate by adding 500 µL of MagMAX™ CORE Wash Solution 2 to each well of a deep well plate (make sure that you label the plate as WASH 2).

11.6 Prepare Elution plate by adding 90 µL of MagMAX™ CORE Elution Buffer to each well of a standard plate (make sure that you label the plate with worklist date).

12. Sample preparation

12.1 Add 450 µl of lysis buffer (step 5.2) to each zirconia bead tube corresponding to a sample.

12.2 Pipet to mix and transfer 200 µl of sample to the zirconia tube in a biological safety cabinet.

12.3 Be sure that all caps are inserted into the tubes to the same depth.

12.4 Place the bead tube rack in the Bead-Ruptor 96 (Omni, cat. #SKU: 27- 0001) using the appropriate lid.

12.5 Bead beat the samples at 20 Hz for 2.5 min. Rest for 5 min, and bead beat for an additional at 20 Hz for 2.5 min. Be sure to switch off and unplug the Bead-Ruptor 96.

12.6 Centrifuge samples at 2,500 rpm for 5 min (Be sure to put the plate rotor correctly).

12.7 Invert the tube of Bead/Proteinase K and vortex to mix several times to resuspend the beads. Then add 30 µl of Bead/Proteinase K mix to the required wells on the deep well plate labelled "Sample".

12.8 Using the decapper (ISC BioExpress, cat. # P-8754-04), remove the caps from the tubes. Discard the caps into the waste container.

12.9 Transfer 500 µl of lysate from bead tubes to the sample plate with bead mix. Be careful not to transfer any zirconia/silica beads. Seal the plate

12.10 Using the ThermoMixer C (Eppendorf, cat. # 2231000574) shake sealed sample plate at 1400 rpm for 2 min.

12.11 Unseal the sample plate and add 350 µl of **MagMAX™ CORE Binding Solution** to each sample well. Immediately proceed to magnetic particle processor (KingFisher Flex).

13. Setting up the equipment

13.1 Turn on the KingFisher Flex. Push the right arrow to select "user" then the down arrow to select "DNA". Toggle down to the "MagMAX_Core_Flex" program and then push start.

13.2 Load each of the plates according to the display's instructions, pressing start after each plate is loaded. Be sure the orientation of the plate is correct (A1 is in the A1 location).

13.3 Press start and record usage in the logbook.

13.4 The program runs approximately 27 minutes. The first plate to be removed is the Elution plate, which now contains the eluted DNA. Place a lid over this plate, label it with the worklist date, and seal it with parafilm. Place the plate with

the eluted DNA on ice. A**l** other plates can be placed in a ziplock bag and discarded in the

biohazard waste, except for the unlabeled plate that held the comb, which can be reused.

13.5 Turn off the KingFisher Flex and wipe down with DNA away solution (Thermo Scientific, cat. # 7000).

13.6 **The elution plate can now be used for bacterial whole genome sequencing methods.**

14. References

Applied Biosystems, Instruction Manual for MagMAX™ CORE Nucleic acid purification kit (cat. #A32700 or A32702)
AAVLD Section 5.4 Test
Methods ISO 17025
Section 5.4 Test
Methods
AHDC QM Section 5.4 Test Methods and Method Validation