

AMR TECHNICAL SCORECARD VETERINARY

Bacterial Culture, Detection,
Identification and Antimicrobial
Susceptibility Testing of Milk
Samples

Milk

Version 1.1 – August 2021

IN PARTNERSHIP WITH

FIND 
Diagnosis for all

ASLM
AFRICAN SOCIETY FOR LABORATORY MEDICINE

Score

Section	Sum of maximum points ¹	Current Audit		Previous audit	
		Date:		Date:	
		Current audit score		Previous audit score	
1. Documents and Records			%		%
2. Management Reviews			%		%
3. Organization and Personnel			%		%
4. Client Management and Customer Service			%		%
5. Equipment			%		%
6. Evaluation and Audits			%		%
7. Purchasing and Inventory			%		%
8. Process Control and Internal and External Quality Assessment			%		%
9. Information Management			%		%
10. Corrective Action			%		%
11. Occurrence Management and Process Improvement			%		%
12. Facilities and Safety			%		%
Milk Module Total			%		%
Milk Module Stars²					

¹ Total number of points of all questions minus points for questions answered with NA.

² No Stars: < 55%
 1 Star: 55% - 64%
 2 Stars: 65% - 74%
 3 Stars: 75% - 84%
 4 Stars: 85% - 94%
 5 Stars: ≥95%

A. General Information

Name of assessor(s)			
Title & organization of assessor			
Name of laboratory being assessed			
Date, type and scope of last assessment?	Date	Type	Score
Internal			
External			
Did the last assessment include assessment of bacterial culture of feces?	Y / N		

B. Technical Information

M-A How many milk cultures and molecular tests were performed last year^{3,4}?

	Milk culture				Molecular ⁵				Clinical diagnosis	Active surveillance
	Q1	Q2	Q3	Q4	Q1	Q2	Q3	Q4	Entire year	Entire year
Farms: Bulk milk										
<i>S. aureus</i>										
<i>S. agalactiae</i>										
<i>S. uberis</i>										
<i>S. dysgalactiae</i>										
<i>C. bovis</i>										
<i>K. pneumoniae</i>										
<i>E. coli</i>										
<i>P. aeruginosa</i>										
<i>Mycoplasma spp.</i>										
Other isolates (specify)										
Other isolates (specify)										
Other isolates (specify)										
Farms: Individual cows										
<i>S. aureus</i>										
<i>S. agalactiae</i>										
<i>S. uberis</i>										
<i>S. dysgalactiae</i>										
<i>C. bovis</i>										
<i>K. pneumoniae</i>										
<i>E. coli</i>										
<i>P. aeruginosa</i>										
<i>Mycoplasma spp.</i>										
Other isolates (specify)										
Other isolates (specify)										
Other isolates (specify)										
Unknown / other⁶										
<i>S. aureus</i>										
<i>S. agalactiae</i>										
<i>S. uberis</i>										
<i>S. dysgalactiae</i>										
<i>C. bovis</i>										
<i>K. pneumoniae</i>										

³ It is highly recommended that assessors obtain the necessary permission to review the laboratory data. However, if assessors are unable to review the laboratory data this question is NOT compulsory for completion of the assessment.

⁴ Refer to the World Organisation for Animal Health (OIE) for further information on OIE listed diseases and other diseases of importance: <https://www.oie.int/en/international-standard-setting/terrestrial-manual/access-online/>

⁵ Molecular tests performed on milk for the detection of bacterial milk pathogens.

⁶ If the laboratory cannot distinguish between samples originating from farms or slaughterhouses, the number of organisms isolated should be recorded as "Unknown/other".

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<i>E. coli</i>										
<i>P. aeruginosa</i>										
<i>Mycoplasma spp.</i>										
Other isolates (specify)										
Other isolates (specify)										
Other isolates (specify)										
TOTAL NUMBER OF ISOLATES										
TOTAL NUMBER OF MILK CULTURES PERFORMED										
TOTAL NUMBER OF CONTAMINATED MILK CULTURES										
TOTAL NUMBER OF NEGATIVE MILK CULTURES										

Q = Quarter

M-B Are there any significant variations (> 20%) in the number of milk culture or molecular tests performed or organisms isolated each quarter? If 'Yes', please explain

Section 1: Documents & Records

All generic requirements apply, see SLIPTA Section 1. In addition, assessors should review the following:

SLIPT			N	Y	P	N	Comments	Score
A			A					
1.5	M1.1	Does the laboratory have documentation covering the following processes?						2
		a) Production of Blood Agar, MacConkey Agar or other media for milk pathogen isolation?						
		b) Microscopic examination and somatic cell count						
		c) Processing of milk culture and molecular tests						
		d) Detection, identification and AST of milk pathogens						
		e) Reporting of milk culture and molecular test results						
		f) Interlaboratory comparison or proficiency testing (PT)						
		g) Laboratory safety						
1.5	M1.2	Are the documents complete, in-date and witnessed by all staff performing milk culture and molecular tests?						2
1.5	M1.3	Are the following processes documented?						3
		a) Rejection criteria for milk samples? ⁸						
		b) How to identify potential pathogens on all primary media? (SOP should describe colony appearance of						

⁷ See ISO15189:2012 Clause 5.5.3 for minimum requirements for a technical Standard Operating Procedure (SOP).

⁸ For more information see OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals 2019 [chapter 1.1.2: Collection, submission and storage of diagnostic specimens](#)

		potential pathogens and define how to proceed when a potential pathogen is encountered)					
		c) Semi-quantitative culture for milk samples?					
		d) Which organisms to identify based on relative quantities (> 10 ⁴ CFU / ml)?					
		e) Instructions for referral of milk culture and molecular tests not performed at the laboratory?					
		f) Instructions for handling samples received after hours?					
		g) Instructions for referral of bacterial isolates for identification and AST?					
		h) Instructions on how to determine AST conversions for automated, disk diffusion, Etest/Gradient and microdilution AST?					
		i) Definition of rare/ unexpected AST results?					
		j) Confirmatory tests for unusual or unexpected patient AST results?					
		k) Turnaround time for milk culture and molecular tests ⁹ ?					
Section 1: Documents & Records Subtotal							7

Section 2: Management Reviews

⁹ From sample collection to reporting.

Section 3: Organization & Personnel

All generic requirements apply, see SLIPTA Section 3. In addition, assessors should review the following:

SLIPT			N	Y	P	N	Comments	Score
A			A					
3.6	M3.1	Is there evidence that laboratory staff have been trained in the following ¹⁰ :						3
		a) Microscopic examination and somatic cell count						
		b) Processing of milk samples for culture and molecular tests						
		c) Detection / identification and AST of milk pathogens						
		d) Interpretation of milk culture and molecular test results						
		e) Reporting of milk culture and molecular test results						
		f) QC for milk culture and molecular tests						
		g) Laboratory safety						
3.7	M3.2	Is there evidence that laboratory staff are following the procedures described in the laboratory documentation? ¹¹ :						3
		a) Microscopic examination and somatic cell count						
		b) Processing of milk samples for culture and molecular tests						
		c) Interpretation of milk culture and molecular test results						
		d) Identification and AST of milk						

¹⁰ Review training records, competency assessment forms and duty rosters. Pay attention to date of training and scope of training compared with techniques being performed.

¹¹ Directly observe procedures being performed compared to the SOP.

		pathogens						
		e) Reporting of milk culture and molecular test results						
Section 3: Organization & Personnel Subtotal								6

Section 4: Client Management & Customer Service

All generic requirements apply, see SLIPTA Section 4. In addition, assessors should review the following:

SLIPT			N	Y	P	N	Comments	Score
A			A					
4.1	M4.1	Is there evidence that the laboratory has provided clients with information/instructions on milk collection, storage and transportation to the laboratory?						3
4.1	M4.2	Is there evidence that the laboratory has provided clients with information/instructions on interpretation of milk microscopy, culture and molecular results and AST?						2
Section 4: Client Management & Customer Service Subtotal								5

Section 5: Equipment

Section 6: Evaluation and Audits

Section 7: Purchasing & Inventory

All generic requirements apply, see SLIPTA Section 7. In addition, assessors should review the following:

SLIPT			N	Y	P	N	Comments	Score
A			A					
7.10	M7.1	Is all media for bacterial culture isolation, identification and AST stored correctly and in date (from date of manufacture media						2

SLIPT		N	Y	P	N	Comments	Score
A		A					
		must be stored at 2-8°C) ¹² ?					
		• Blood Agar					
		• MacConkey agar					
		• M-17 or equivalent					
		• Mueller Hinton					
Section 7: Purchasing & Inventory Subtotal							2

Section 8: Process Control

All generic requirements apply, see SLIPTA Section 8. In addition, assessors should review the following:

SLIPT		N	Y	P	N	Comments	Score	
A		A						
SPECIMEN COLLECTION								
8.5	M8.1	If milk samples will reach the laboratory more than 2 hours post collection, are they transported to the laboratory on ice?					2	
MEDIA QUALITY CONTROL								
8.8	M8.2	Does the laboratory perform QC testing on all media before use ¹³ ?					3	
		Blood agar						
		Do QC records for blood agar plates demonstrate that they are checked for their ability to support growth of fastidious organisms such as <i>S. pneumoniae</i> ?						
		Do QC records for blood agar plates demonstrate that they are checked for their ability to show beta, alpha, and gamma hemolysis?						
		MacConkey agar (MAC)						
		Do QC records for MAC plates demonstrate that they are checked for their ability to suppress growth of Gram -positive organisms while						

¹² According to manufacturer's requirements.

¹³ This includes in-house made or purchased from commercial sources.

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SLIPT			N	Y	P	N	Comments	Score
A			A					
		allowing the growth of Gram -negative organisms?						
		Do QC records for MAC plates demonstrate that they are checked for their ability to allow visualization of lactose fermentation?						
M-17 or equivalent								
		Do QC records for M-17 or equivalent agar plates demonstrate their ability to support lactic streptococci?						
Mueller Hinton Agar (MHA)								
		Do QC records demonstrate that MHA plates are checked for their ability to grow <i>S. aureus</i> & <i>E. coli</i> ?						
8.8	M8.3	Does the laboratory:						
		a) Perform sterility and performance tests for every batch of culture media using certified reference strains as controls?						
		b) Source their reference strains from an authorized supplier (e.g. ATCC)?						3
		c) Store, culture and sub-culture the reference strains in accordance with the specification from the supplier?						
8.10	M8.4	Does the laboratory determine the cause of failed QC (root cause analysis), perform corrective actions and measure their effectiveness?						2
BACTERIAL MILK CULTURE PROCEDURE								
8.7	M8.5	Are all milk samples processed within 2 hours of collection, or a						2

SLIPT A			N A	Y	P	N	Comments	Score
		maximum of 4 hours after collection if transported on ice?						
8.7	M8.6	Does the laboratory perform a somatic cell count on all milk specimens prior to inoculation on culture media in order to determine the number of somatic cells in the milk?						2
8.7	M8.7	Does the laboratory have a procedure for rechecking somatic cell counts to ensure consistency of microscopic observations/interpretations among all personnel performing microscopy?						2
8.7	M8.8	Does the laboratory perform a bacterial culture on all milk samples (or those with somatic cell counts $>10^5$ white blood cells/mL) as per their policy?						2
8.7	M8.9	Is milk plated using a calibrated 1 μ L loop?						2
8.7	M8.10	Are the following media used for primary culture of milk? <ul style="list-style-type: none"> • Blood Agar or equivalent • MacConkey Agar or equivalent • M-17 or equivalent¹⁴ 						2
8.7	M8.11	Are media used for primary culture of milk incubated aerobically at 35-37°C for at least 18 hours?						2
8.7	M8.12	Does the lab use appropriate criteria for determining						2

¹⁴ M-17 Agar media is proposed for the cultivation, detection and enumeration of lactic streptococci in milk and dairy products. The M-17 media are superior to other comparable culture media for the cultivation of the fastidious species *Streptococcus cremoris*, *Streptococcus diacetylactis* and *Streptococcus lactis*.

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SLIPT		N	Y	P	N	Comments	Score
A		A					
		contamination of a milk culture specimen? (polymicrobial culture/no predominant colonies >10 ⁴ CFU)					
BACTERIAL ID & AST							
8.7	M8.13	Does the laboratory perform identification tests (ID) for at least the following milk pathogens? <ul style="list-style-type: none"> • <i>S. aureus</i> • <i>S. agalactiae</i> • <i>S. uberis</i> • <i>S. dysgalactiae</i> • <i>C. bovis</i> • <i>K. pneumoniae</i> • <i>E. coli</i> • <i>P. aeruginosa</i> • <i>Mycoplasma spp</i> 					2
8.7	M8.14	Does the laboratory perform AST on at least the following milk pathogens using an approved test method? <ul style="list-style-type: none"> • <i>S. aureus</i> • <i>S. agalactiae</i> • <i>S. uberis</i> • <i>S. dysgalactiae</i> • <i>C. bovis</i> • <i>K. pneumoniae</i> • <i>E. coli</i> • <i>P. aeruginosa</i> • <i>Mycoplasma spp</i> 					2
8.7	M8.15	Is the following testing performed for <i>S. aureus</i> identification? ¹⁵ <ul style="list-style-type: none"> • Catalase • Coagulase (slide or tube) • Mannitol Salt Agar (MSA) • Dnase 					2

¹⁵ If the laboratory performs penicillin AST, it is recommended that *S. aureus* isolates with penicillin zones sizes or MICs in the susceptible range are tested for B-lactamase production using the zone-edge test or a nitrocefin test before being reported as penicillin susceptible.

SLIPT A			N A	Y	P	N	Comments	Score
8.7	M8.16	Does <i>S. aureus</i> AST include the following antibiotics ¹⁶ : <ul style="list-style-type: none"> • Cefoxitin • Vancomycin 						2
8.7	M8.17	Does the laboratory detect methicillin/nafcillin resistance in <i>S. aureus</i> using oxacillin disk?						2
8.7	M8.18	Is the following testing performed for <i>Streptococcus sp.</i> identification? <ul style="list-style-type: none"> • Bacitracin • Pyrrolidonyl Arylamidase (PYR) • Bile solubility • Catalase • Optochin 					2	
8.7	M8.19	Does <i>Streptococcus sp.</i> AST include the following antibiotics: <ul style="list-style-type: none"> • Oxacillin • Co-trimoxazole • Ceftriaxone or cefotaxime 					2	
8.7	M8.20	Is the following testing performed for <i>Corynebacterium sp.</i> identification? <ul style="list-style-type: none"> • Catalase • Nitrate reduction • Lipid dependency 					2	
8.7	M8.21	Does <i>Corynebacterium sp.</i> AST include the following antibiotics: <ul style="list-style-type: none"> • Penicillin • Erythromycin • Tetracycline 					2	

¹⁶ If oxacillin and cefoxitin results are discrepant for *S. aureus* (one is susceptible and one is resistant), the laboratory should repeat the testing. Note: oxacillin testing should always be tested by MIC (not disc diffusion). If the results remain discrepant, oxacillin should be reported as resistant.

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SLIPT A			N A	Y	P	N	Comments	Score
8.7	M8.22	Is the following testing performed to identify Gram negative bacilli? <ul style="list-style-type: none"> • Oxidase • Indole • Methyl Red • Voges Proskauer • Citrate • Triple Sugar Iron or Kligler Iron • Urease • Motility 						2
8.7	M8.23	Is Gram negative bacteria AST done as per current CLSI Vet/ VetCAST guidelines for diagnostic testing and CLSI/EUCAST guidelines for surveillance testing? ¹⁷						2
8.7	M8.24	Does the laboratory test for <i>Mycoplasma spp.</i> using molecular methods?						2
8.7	M8.25	Does the laboratory use Combination Disk Test or another equivalent method for Extended Spectrum Beta-Lactamase (ESBL) screening? ^{18,19?}						2
8.7	M8.26	Does the laboratory use Combination Disk Test or another equivalent method for carbapenemase screening?						2
INTERLABORATORY COMPARISON, PT AND EXTERNAL QUALITY ASSURANCE (EQA)								
8.14	M8.27	Is the laboratory enrolled in an interlaboratory comparison and/or PT program for milk culture and/or molecular tests for organism identification and AST?						2

¹⁷ See user guide section 3.4.5 for links to CLSI, EUCAST, CLSI veterinary and VetCAST guidelines.

¹⁸ J Clin Microbiol. 2013 Sep; 51(9): 2986–2990.

¹⁹ https://www.eucast.org/fileadmin/src/media/PDFs/EUCAST_files/Resistance_mechanisms/EUCAST_detection_of_resistance_mechanisms_170711.pdf

SLIPT A			N A	Y	P	N	Comments	Score
8.14	M8.28	Did the laboratory pass the last 3 rounds of interlaboratory comparison or PT program testing?						2
8.14	M8.29	Does the laboratory receive onsite supervision visits as part of the EQA program for milk culture and/or molecular tests?						2
Section 8: Process Control Subtotal								60

Section 9: Information Management

All generic requirements apply, see SLIPTA Section 9. In addition, assessors should review the following:

SLIPT A			N A	Y	P	N	Comments	Score
9.3	M9.1	Does the final report for milk culture list the organisms for which the specimen was and was not cultured ²⁰ ?						2
9.3	M9.2	Does the laboratory report alert organisms which include at least the following: ²¹ <ul style="list-style-type: none"> • ESBL producing organisms • Methicillin-resistant <i>S. aureus</i> (MRSA) • Colistin Resistant <i>E. coli</i> • Vancomycin resistant <i>S. aureus</i> 					2	
Section 9: Information Management Subtotal								4

Section 10: Identification of Non-conformities, Corrective and Preventive Actions

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²⁰ The laboratory should inform the veterinarian on the report what organisms were excluded during the culture process. This may be either by choice of media or incubation conditions (e.g. anaerobic organisms). Assessors should review a number of laboratory reports to determine how results are reported. Procedures should be consistent with the laboratory's SOPs.

²¹ Alert organisms are organisms with significant public health threat and/or organisms that are notifiable.

Section 11: Occurrence/Incident Management & Process Improvement

All generic requirements apply, see SLIPTA Section 11. In addition, assessors should review the following:

SLIPTA			N	Y	P	N	Comments	Score
A			A					
11.4 & 11.5	M11.1	Are the following performance indicators collected ²² ?						3
		<ul style="list-style-type: none"> Number of milk culture and molecular tests performed (disaggregated by type) <ul style="list-style-type: none"> Farms: Bulk milk Farms: Individual cows Unknown/referred²³ 						
		<ul style="list-style-type: none"> Number and percentage of samples for bacterial milk culture or molecular tests rejected (disaggregated by reason e.g. leaked, insufficient volume) (target <1%) 						
		<ul style="list-style-type: none"> Number and percentage of milk cultures with somatic cell counts > 10⁵ cells/ml 						
		<ul style="list-style-type: none"> Number of milk culture and molecular tests where pathogens were identified/isolated (disaggregated by type) <ul style="list-style-type: none"> <i>S. aureus</i> <i>S. agalactiae</i> <i>S. uberis</i> 						

²² It may not be possible for laboratories to determine the origin of the milk samples if this is not collected on the laboratory requisition form.

²³ If the laboratory can't distinguish the origin of the milk samples, the number of organisms isolated should be recorded as "Unknown/referred".

SLIPT		N	Y	P	N	Comments	Score	
A		A						
	<ul style="list-style-type: none"> ○ <i>S. dysgalactiae</i> ○ <i>C. bovis</i> ○ <i>K. pneumoniae</i> ○ <i>E. coli</i> ○ <i>P. aeruginosa</i> ○ <i>Mycoplasma spp</i> 							
	<ul style="list-style-type: none"> • Number and percentage of contaminated milk culture tests 							
	<ul style="list-style-type: none"> • Milk culture and molecular test TAT²⁴ 							
Section 11: Occurrence/Incident Management & Process Improvement Subtotal								3

Section 12: Facilities and Biosafety

²⁴ From sample collection to reporting.



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