Uricult® Vet
CLED EMB

Instructions for use
36162-3

Intended use
A dip paddle culture method for diagnosing urinary tract infections. Uricult Vet is intended for veterinary use only.

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Principle
The Uricult Vet dip paddle system is based on two culture media. One side of the plastic paddle is covered with green CLED media and the other with reddish EMB media for the detection of microbes causing urinary tract infections. The CLED media is intended for determining the total microbial count. The EMB media is intended for detecting gram-negative* microbes.

Typical formulation

CLED media		**EMB media**	
Peptone	8.0 g/l	Peptone	10.0 g/l
Meat extract	3.0 g/l	Lactose	5.0 g/l
Lactose	10.0 g/l	Sucrose	5.0 g/l
L-Cystine	0.13 g/l	Dipotassium Phosphate	2.0 g/l
Bromthymol blue		Eosin Y	
		Methylene Blue	

Warnings and precautions
- Uricult Vet is for ***in vitro*** **diagnostic use** only.
- Do not use the product beyond the expiration date marked on the box.
- Do not use the product if you detect discoloration or dehydration of the culture media, separation of the culture media from the plastic paddle or evidence of microbial growth on the culture media.
- Because any colonies growing on the Uricult Vet culture media are actual or potential pathogens, do not touch the growth.
- To avoid contamination**, do not touch the surfaces of uninoculated Uricult Vet culture media.
- Ensure that the surfaces of the culture media do not come into contact with animal hair or other objects in conjunction with sampling.

Storage
Uricult Vet is stored at room temperature (18...25°C / 64...77°F), protected from air and temperature fluctuations. A small amount of condensed water (<0.5 mL) may accumulate on the bottom of the tube during storage. This water does not affect the performance of the test nor does it shorten the shelf-life.
Avoid drafts and storage near heat-generating appliances. DO NOT ALLOW TO FREEZE. The expiration date is marked on the box.

Sampling
Ideally, urine for bacterial culture should remain in the bladder for four hours prior to sampling. The veterinarian can take a sample via catheterization or cystocentesis as required.
The urine sample may also be taken by holding a Uricult Vet paddle directly in the animal's urine stream or in a collection cup. **Please note that this sampling method is not preferred.** In this method, it is important ensure that the surfaces of both culture media become completely wet and that no contamination occurs from animal hair or other objects in conjunction with sampling.
The urine should be inoculated onto the Uricult Vet paddle immediately after collection. The paddle should then at once be returned into its protective tube and the cap closed.

A paddle that has been dipped in the sample may be
a) incubated immediately or
b) stored at 2...8°C / 36...46°F for up to 48 hours or
c) transported to a laboratory for incubation and/or interpretation

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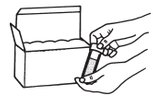
Test procedure

1



Unscrew the paddle from the tube without touching the surfaces of the culture media.

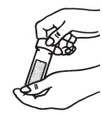
2

Holding Uricult Vet by the cap, dip the paddle into the urine sample so that the surfaces of the culture media become completely wet. If the volume of urine is too small for this, the culture media can be inoculated by pouring urine on them while tilting the paddle from side to side to ensure complete wetting.

3

Allow excess urine to drain from the Uricult Vet paddle. The base of the culture-paddle may be blotted on absorbent paper if desired.

4



Return the paddle into the tube and close the tube.

5

Fill in the patient label and attach it to the tube.

6

To incubate*** Uricult Vet, place the tube upright in an incubator (36°C ± 2°C / 97°F ± 4°F) for 16–24 hours. Uricult Vet may also be sent to a laboratory for incubation and interpretation.

7

Interpret the results.

Contact information

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ISO9001 CERTIFIED ISO13485

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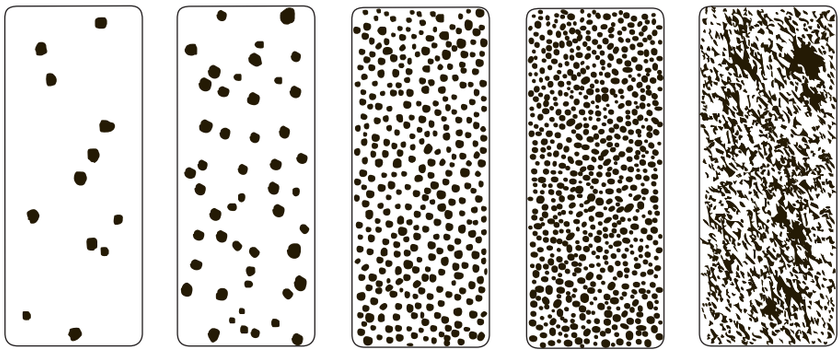
Colony Density Chart



CFU/ml	1 000	10 000	100 000	1 000 000	10 000 000
	10^3	10^4	10^5	10^6	10^7

The colony count is determined from the originally green CLED media by matching the colony density with the model chart it most closely resembles. Low volume of urine may cause it to spread unevenly on the different media. In these cases, it is advisable to evaluate the growth on both media. If there is a significant difference in the number of colonies on each side, the side with the greater number should be used for determining the colony count.
It is important to compare the number of colonies, not their size.
A growth consisting of several species of bacteria is termed mixed flora and is most likely due to contamination of the urine sample.

Interpretation of results

Remove the Uricult Vet paddle from the incubator following incubation period. Compare colony count density on the media surfaces with the Colony Density Chart provided to obtain a semiquantitative colony count in CFU/ml of urine. Compare only the number of colonies present, not the size of the colonies on the media surface area they cover. The colonies on the agar may also be observed at this time for the morphology and the media color reactions which may also be used for presumptive identification of the bacterial growth.

Note:
Uricult Vet cultures that appear negative after incubation, may be incubated for additional 24 hours at 36°C ± 2°C / 97°F ± 4°F to detect any slow-growing bacteria and yeasts.

Confluent growth
This can be interpreted as a negative result. Therefore, any culture media surfaces that appear negative should be examined under a reflecting light; absence of reflection suggests confluent growth. A bright light also facilitates the detection of pinpoint colonies.
A change in color of the CLED media is also an indication of growth.
A growth consisting of several species of bacteria is termed mixed flora and is most likely due to contamination of the urine sample.

Expected values

Method of sampling	Significant colony count CFU/ml	
	Dog	Cat
Bladder aspiration	≥ 1 000	≥ 1 000
Catheterization	≥ 10 000	≥ 1 000
Voided urine	≥ 100 000	≥ 10 000

Presumptive identification

Staphylococcus aureus:
Gram Positive
Growth of yellow colonies with a color change towards yellow on the CLED media.

Enterococcus faecalis:
Gram Positive
Growth of yellow colonies with a color change towards yellow on the CLED media and with a growth of pin point colonies on the EMB media.

Proteus vulgaris:
Gram Negative
Growth of translucent colonies with a color change towards blue on the green CLED media. Growth of colorlesss colonies on the EMB media.

Escherichia coli:
Gram Negative
Growth of yellow colonies with a color change towards yellow on the CLED media and growth of purple or metallic green colonies on the EMB media.

Gram (+) or (–)			
Organism	**CLED**	**EMB**	**Gram**
S. aureus	G	TNG	+
E. faecalis	G	G	+
P. vulgaris	G	G	–
E. coli	G	G	–

G = Growth TNG = Typically No Growth NG = No growth

Limitations of procedure

Uricult Vet is capable of detecting urinary bacterial concentrations between 10^3 and 10^7 CFU/ml. The colony chart allows the determination of colony counts to the nearest power of 10. When the method is used according to instructions, the colony counts show a 99% correlation with the conventional pour plate method.
Antibiotic medications may affect the result of the Uricult Vet test. Therefore, the test should not be performed until 48 hours after the final dose of medication.
Storage or transportation should not exceed 48 hours at 7...25°C / 45...77°F, after which Uricult Vet should be incubated at 36°C ± 2°C / 97°F ± 4°F. If Uricult Vet has been stored or transported for 48 hours, only the presence of growth and the colony count may be recorded from it; the color reaction of the culture media may be atypical. Voided urine specimens may contain bacteria contaminants from the animals skin or fur.

Performance characteristics

CLED media
Arneil, G.C. 1970: Detection of bacteriuria at room temperature: Lancet, January 17, pp 119–121.

Number of samples	140
Sensitivity	100%
Specificity	99%
PPV	98%
NPV	100%
Reference method	Pour plate

Quality assurance

Quality assurance tests are performed on each lot of Uricult Vet dip paddles at the time of manufacture. Should the user wish to perform his/her own quality assurance, the following procedure is recommended:

1. Prepare a 10^5–10^6 bacteria/ml suspension of each of the following bacterial species in sterile saline:
   a. *Staphylococcus aureus* ATCC 25923
   b. *Escherichia coli* ATCC 25922
   c. *Proteus mirabilis* ATCC 12453
2. Use the suspensions to inoculate Uricult Vet dip paddles, using the normal procedure.

Disposal

Used Uricult Vet dip paddles should be disposed of **with adherence to local biohazard regulations.**

06 PS	05 PP	06 PS	04 PE-LD
Tube, clear	Tube, mat	Paddle	Cap

Glossary

* **gram-positive and gram-negative**:
Gram staining is the most important method of grouping bacteria, allowing bacteria to be classified either as gram-positive (staining blue) or gram-negative (staining red). The differential staining is due to differences in the structure of the bacterial cell wall.

** **Contamination** denotes the presence of microbes in the urine that were introduced by the sampling procedure.

*** **Incubation** denotes growing of Uricult Vet cultures in an incubator at 36 ± 2ºC.

**** **Lactose-positive** denotes a bacterium capable of utilizing/fermenting the lactose contained in the culture media.
Lactose-negative denotes a bacterium incapable of utilizing/fermenting the lactose contained in the culture media.