

# MACCONKEY AGAR w/ 1 µg/ml CIPROFLOXACIN

## INTENDED USE

Remel MacConkey Agar w/ 1µg/ml Ciprofloxacin is a solid medium recommended for use in qualitative procedures for primary, selective isolation of fluoroquinolone-resistant enteric gram-negative bacilli in clinical specimens.

## SUMMARY AND EXPLANATION

Ciprofloxacin is a broad-spectrum, fluoroquinolone antibiotic introduced for clinical use in the 1980s.<sup>1</sup> It has been used extensively in the decades since due to its broad antimicrobial spectrum, availability in both oral and parenteral formulations, and low toxicity profile. Ciprofloxacin is frequently used empirically to treat infections with organisms that may be susceptible to other antibiotics and for prophylactic therapy of patients prior to certain invasive procedures, such as transrectal ultrasound-guided biopsy. Fluoroquinolone resistance among gram-negative bacilli began to emerge in the 1990s.<sup>2</sup> A national review of ICU patients in 43 states during 1994-2000 found the overall susceptibility to ciprofloxacin had decreased from 86% to 76%, which correlated with the greater than 2.5-fold increase in fluoroquinolone use during the same time period.<sup>3-5</sup>

In the United States, transrectal ultrasound-guided biopsy is the standard procedure for the diagnosis of prostate cancer.<sup>6</sup> Infectious complications caused by fluoroquinolone-resistant *Escherichia coli* following prostate biopsy have increased substantially in recent years.<sup>7</sup> In 2011, Liss et al. proposed a method to identify patients colonized with even small numbers of fluoroquinolone-resistant *E. coli*.<sup>8</sup> Rectal swab specimens, collected immediately prior to biopsy, were placed in brain heart infusion broth supplemented with ciprofloxacin. After overnight incubation each broth was subcultured to a MacConkey agar plate. Fluoroquinolone-resistant *E. coli* was isolated from 22% of the patients in this study. In 2012, Taylor et al. reported the results of a study using targeted prophylaxis for men undergoing transrectal ultrasound-guided prostate biopsy.<sup>9</sup> A total of 457 men were included in the study; 112 men (24.5%) had rectal swabs obtained at least 5-7 days prior to biopsy and the remaining 345 patients received standard empirical prophylaxis. Rectal swab specimens were inoculated onto a selective medium containing ciprofloxacin. Patients identified as colonized with fluoroquinolone-resistant organisms received targeted antimicrobial prophylaxis rather than empirical treatment with ciprofloxacin. None of the men receiving targeted prophylaxis had infectious complications, including those (19.6%) colonized with fluoroquinolone-resistant organisms. In contrast, 9 patients who received empirical prophylaxis had infectious complications; 7 with fluoroquinolone-resistant organisms and 2 with fluoroquinolone-susceptible organisms.

MacConkey Agar w/ 1µg/ml Ciprofloxacin is a selective agar for the isolation of ciprofloxacin-resistant gram-negative bacilli. It is intended for use following pre-enrichment of the specimen in BHI Broth w/ 1µg/ml Ciprofloxacin. The use of MacConkey Agar w/ 1 µg/ml Ciprofloxacin in combination with BHI Broth w/ 1µg/ml Ciprofloxacin to identify patients colonized with fluoroquinolone-resistant organisms allows for the use of targeted antibiotic therapy, helps to avoid treatment failures, and prevents further spread of ciprofloxacin-resistant organisms.

## PRINCIPLE

Peptones provide nitrogen compounds and amino acids necessary for bacterial growth. Sodium chloride is a source of essential electrolytes and maintains osmotic equilibrium. Crystal violet and bile salts are selective agents which inhibit most gram-positive organisms. Differentiation of gram-negative bacilli is accomplished by addition of lactose and neutral red which is an indicator. Gram-negative bacilli which ferment lactose form pink colonies. Lactose nonfermenters form colorless, transparent colonies. Ciprofloxacin is an antibiotic which provides for selective isolation of fluoroquinolone-resistant gram-negative bacilli. Agar is a solidifying agent.

## REAGENTS (CLASSICAL FORMULA)\*

Gelatin Peptone.....	17.0 g	Meat Peptone.....	1.5 g
Lactose .....	10.0 g	Neutral Red.....	30.0 mg
Sodium Chloride.....	5.0 g	Ciprofloxacin .....	10.0 mg
Bile Salts .....	1.5 g	Crystal Violet.....	1.0 mg
Casein Peptone.....	1.5 g	Agar .....	13.5 g
		Demineralized Water .....	1000.0 ml

pH 7.1 ± 0.2 @ 25°C

\*Adjusted as required to meet performance standards.

## PROCEDURE

1. Place the swab in BHI Broth w/ 1µg/ml Ciprofloxacin (R060274) as soon as possible after it is received in the laboratory.
2. Incubate the tube at 33-37°C for 18-24 hours with cap loosened.
3. Transfer 0.1 ml of the broth to a plate of MacConkey Agar w/ 1µg/ml Ciprofloxacin (R01545) and streak for isolation.
4. Incubate the plate in ambient air, protected from light, at 33-37°C for 18-24 hours. If no growth is observed at 24 hours, reincubate the plate for an additional 24 hours.
5. Examine plate for typical colony morphology. Lactose-fermenters form pink colonies and nonlactose-fermenters form colonies which are transparent or colorless.
6. All colonies require further testing (e.g., biochemical, etc.) for definitive identification. Confirmatory antimicrobial susceptibility testing must be performed on all isolates according to established laboratory procedures. Consult appropriate references for further instructions.<sup>1,10</sup>

## QUALITY CONTROL

All lot numbers of MacConkey Agar w/ 1 µg/ml Ciprofloxacin have been tested using the following quality control organisms and have been found to be acceptable. Testing of control organisms should be performed in accordance with established laboratory quality control procedures. If aberrant quality control results are noted, patient results should not be reported.

## CONTROL

*Acinetobacter baumannii* BAA-1605  
*Escherchia coli* ATCC® 25922

## INCUBATION

Ambient, up to 48 h @ 33-37°C  
Ambient, up to 48 h @ 33-37°C

## RESULTS

Growth, slightly pink to colorless colonies  
Inhibition, partial to complete

## LIMITATIONS

1. MacConkey Agar w/ 1 µg/ml Ciprofloxacin is recommended for selective isolation of gram-negative bacilli with increased resistance to fluoroquinolone antibiotics. It is not intended for use as a method of antimicrobial susceptibility testing.
2. Confirmation of resistance by an approved method is necessary, as some organisms on initial isolation may overcome the inhibitory effects of this medium.
3. The absence of suspect colonies on MacConkey Agar w/ 1 µg/ml Ciprofloxacin does not rule out the presence of fluoroquinolone-resistant organisms.

## BIBLIOGRAPHY

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Refer to the front of Remel *Technical Manual of Microbiological Media* for General Information regarding precautions, product storage and deterioration, specimen collection, storage and transportation, materials required, quality control, and limitations.

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IFU 1545, Revised August 20, 2013

Printed in U.S.A.

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