

Several mechanisms for the development of gastric dilatation in anorexia nervosa have been suggested. In this girl, in three other reported cases,²⁻⁴ and in emaciated prisoners of war, recounted from Markowski by Russell,⁵ dilatation occurred soon after a sudden increase in normal food intake. Scobie³ reports duodenal dilatation as common in anorectic patients undergoing barium meal examination for causes other than gastric dilatation. He suggests that this is a neurogenic complication of malnourishment. Possibly this functional duodenal obstruction exacerbated by a sudden load on the stomach may produce dilatation. This supposition is supported by the presence of upper gastrointestinal tract dilatation in association with some cases of gastric dilatation.²⁻⁴ In our case and in Russell's there was no evidence of electrolyte disturbance as a cause for the dilatation. Russell⁵ suggested that psychogenic factors were important, but other authors¹⁻⁴ have not reported them nor could we find any in our patient. Drugs may contribute to this complication, but, although most patients have been treated with phenothiazines, one¹ was receiving only diazepam. We have been unable to find other reports of gastric dilatation associated with diazepam. Jennings and Klidjian⁴ quote an incidence of four cases of undefined "paralytic ileus" occurring in 720 patients treated with phenothiazines, but there is no specific mention of gastric dilatation in reports on phenothiazine side effects.

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¹ Evans, D S, *British Journal of Surgery*, 1968, **55**, 940.

² Di Costanzo, J, *et al*, *La Nouvelle Presse Médicale*, 1975, **4**, 590.

³ Scobie, B A, *Medical Journal of Australia*, 1973, **2**, 932.

⁴ Jennings, K P, and Klidjian, A M, *British Medical Journal*, 1974, **2**, 477.

⁵ Russell, G F M, *British Journal of Psychiatry*, 1966, **112**, 203.

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Penicillinase-producing *Neisseria gonorrhoeae*: detection by starch paper technique

A recent editorial in the *Lancet*¹ drew attention to the need for study of the distribution of penicillinase-producing gonococci in different parts of the world to determine whether they are merely freaks or whether they have become firmly established in various populations. As these organisms may constitute a worldwide problem, some simple, reliable, and inexpensive method for the detection of penicillinase production is essential. We report here a modification of the iodometric technique,² a simple method which would place the detection of penicillinase-producing strains within the reach of any clinical bacteriology laboratory.

Materials, methods, and results

Benzylpenicillin solution containing 100 000 µg/ml in phosphate-buffered saline (PBS) Oxoid, pH 7.3 approx. Iodine solution: Gram's iodine diluted 1 in 2. Starch paper: any good white bond paper (for example, Basildon Bond) is tested for the presence of starch; satisfactory samples of paper turn blue-black on the addition of a drop of iodine solution. The paper is cut into 7 cm × 4 cm strips to fit the bottom of Petri dishes.

Organisms—(a) Beta-lactamase-positive controls: *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumoniae*, *Enterobacter* spp, *Bacillus* spp. (b) Beta-lactamase-negative controls: The Oxford strain of staphylococcus, and *Neisseria gonorrhoeae* strain F62. (c) 100 strains of *N gonorrhoeae* including two known beta-lactamase producers. The identity of all the strains of *N gonorrhoeae* tested was confirmed by standard methods.

Procedure—The strips of starch paper were soaked for 10 minutes in the solution of benzylpenicillin and then spread smoothly in a Petri dish. Each strip of paper was used to test six organisms including the controls. With a fine bacteriological loop (2 mm diameter), colonies of bacteria were collected from the surface of the culture plate and then transferred to the surface of the test paper and spread over an area of 2-3 mm; the inocula were placed at least 1.5 cm apart. The plates were incubated at 37°C for 30 minutes,

after which the paper was flooded with the iodine solution (which was drained off immediately); this causes the paper to turn uniformly black within about 30 seconds.

Subsequently areas where penicillin is *inactivated*—for example, immediately around the area of application of the organism—are decolorised and contrast with the remaining part, which is blue-black. This is because the starch in the bond paper combines with iodine to form blue-black starch-iodine complex, and, where beta-lactamase (penicillinase) is present, it results in the enzymatic degradation of the beta-lactam ring of penicillin—with the production of penicilloic acid, which reacts with iodine to cause dissociation of the starch-iodine complex.^{2,3}

Penicillinase-producing strains of *N gonorrhoeae* were detected by the decoloration of the blue-black colour surrounding the organisms. The white halo widens in the course of the ensuing five minutes, while the surface of the inoculum remains whitish. Penicillinase-negative gonococci did not produce any decoloration of the surrounding area and the organisms retained a yellow tinge due to unchanged iodine. Results are read within five minutes, after which the background tends to decolorise, making interpretation more difficult. The use of a lower initial concentration of penicillin results in weaker reactions; this confirms observations that weak penicillinase-producing strains of *Staph aureus* are more readily detected in the presence of a high substrate concentration.⁴

The table compares the results obtained using the starch paper method with those of the chromogenic cephalosporin test.⁵

Comparison of beta-lactamase detection by starch paper method and chromogenic cephalosporin test

Organisms	Total No tested	Positive with starch paper method	Positive with chromogenic cephalosporin
<i>Neisseria gonorrhoeae</i> ..	100	2	2
<i>Staphylococcus aureus</i> ..	10	10	10
<i>Escherichia coli</i> ..	5	5	5
<i>Salmonella</i> spp ..	5	0	0
<i>Klebsiella</i> spp ..	3	2	1
<i>Enterobacter</i> spp ..	3	1	1
<i>Bacillus</i> spp ..	3	1	1
<i>Shigella</i> spp ..	3	0	0

Discussion

The emergence of penicillinase-producing gonococci has highlighted the problem of widespread and indiscriminate use of antibiotics, especially in the underdeveloped countries. Thus there is more than ever a need for some simple, reliable, and cheap method for detecting penicillinase-producing organisms; undoubtedly the use of chromogenic cephalosporin allows rapid detection of beta-lactamase by the development of a distinctive red colour,⁵ which has been successfully used to detect beta-lactamase-producing gonococci. This compound is not readily available and would certainly not be found in small clinical laboratories in the tropical or subtropical areas.

The present technique will rapidly detect penicillinase production by *N gonorrhoeae*. As the materials for the test are cheap and readily available, and in our experience the two tests give strikingly similar results with all the bacterial penicillinases we tested, the use of this method should encourage further studies on the prevalence of penicillinase-producing gonococci in the tropics.

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¹ *Lancet*, 1976, **2**, 725.

² Workman, R C, and Farrar, E W, *Journal of Infectious Diseases*, 1970, **121**, 433.

³ Fleming, P C, personal communication.

⁴ Novick, R P, and Richmond, M H, *Journal of Bacteriology*, 1965, **90**, 467.

⁵ O'Callaghan, C H, *et al*, *Antimicrobial Agents and Chemotherapy*, 1972, **1**, 283.

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