A Plate Test with Oxacillin and Penicillinase

Oxacillin, or 5-methyl-3 phenyl-4 isoxazolil penicillin, belongs to the group of penicillins resistant to penicillinase.

Resistance to penicillin G as shown clinically by staphylococci is due to the penicillinase produced in large quantities by these resistant micro-organisms[1].

To check the effect of penicillinase on penicillin G as compared with Oxacillin[2], the following experiment was carried out.

Material and Method. Place 20 ml 'Biosat-Agar' (BBL) in a 10 cm Petri dish. Allow to solidify. With a sterile scalpel cut the agar along a diameter and slide off one half.

Add 1 ml of penicillinase, 50,000 u/ml (BBL) to 10 ml of 'Biosat-Agar' dissolved and cooled to 50°C; shake well and place in the empty part of the dish, filling to the level of the other half. The result is a dish of agar, one half with penicillinase and one half without (the penicillinase diffuses very little into the agar).

Seed whole surface (streaking technique) with a staphylococcus sensitive to penicillin G. Place a disc with sodium penicillin G (2 μg) and a disc with Oxacillin (2 μg) at two points on the dividing line between the two semicircles of the agar surface (with and without penicillinase). Place in incubator at 37°C and read after 20 h.

Results. As may be seen from the Figure, the disc with Oxacillin (P12) produces a perfectly circular inhibition zone. The disc with penicillin G (P) produces a semicircular inhibition zone only on the part of the agar without penicillinase; on the part with penicillinase the growth around the disc is equal to the rest of the surface of the dish.

Discussion. The plate test with two zones of agar, with and without penicillinase, demonstrates the resistance of Oxacillin to penicillinase. The inhibition zone around the P12 disc forms a perfect circle, showing that a concentration of 5000 U penicillinase per ml of agar does not modify the activity of Oxacillin discs. The inhibition zone around the disc with penicillin G, showing a semicircle situated on the side of the agar without penicillinase, demonstrates the effectiveness of penicillin G against the staphylococcus tests; but the growth around the disc of penicillin on the part where penicillinase was added to the agar shows total destruction of the penicillin G.

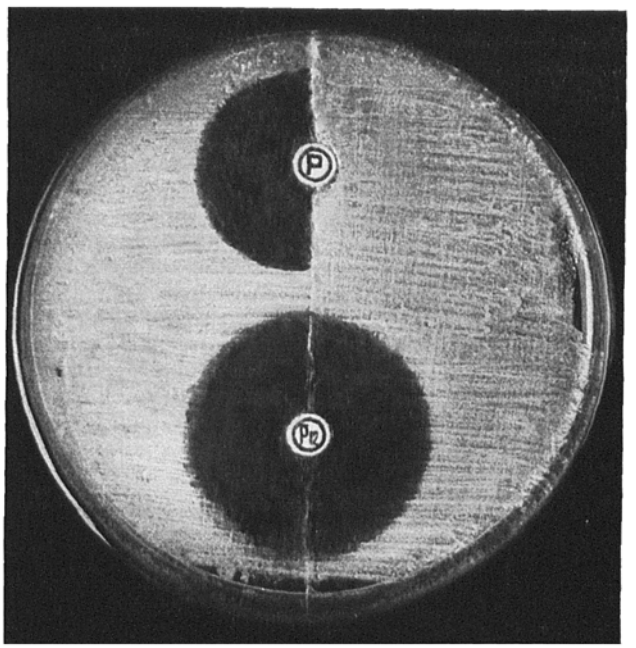

Plate test with two zones of agar, right with, and left without, penicillinase, showing inactivation of the penicillin (disc P, above) and the circular zone produced by Oxacillin (disc P12, below).

Résumé. La résistance de la penicilline P-12 à la penicillinase a été démontrée grâce à une technique sur plaque à deux zones d'agar, avec et sans penicillinase.

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[1] M. Barber, *Drug Resistance of Staphylococci with Special Reference to Penicillinase Production*, Ciba Foundation Symposium on Drug Resistance (London 1957), p. 262.

[2] The Oxacillin was kindly supplied by Laborterapica Bristol; the trade mark for Oxacillin is Prostaphlin.

Immunoélectrophorèse par application directe de l'antigène à la surface du gel

Depuis la description par Grabar et Williams de l'immunoélectrophorèse[1], d'intéressants perfectionnements et diverses variantes de cette méthode ont été proposés[2-8].

L'objet de cette note est de montrer certains avantages d'une technique d'application directe, à la surface du gel, de l'antigène (ou éventuellement de l'anticorps) dont on veut obtenir la résolution électrophorétique.

Le réservoir de départ étant supprimé, la couche de gélose ne présente aucune solution de continuité; ce qui favorise, lors de la migration, une répartition plus uniforme du champ électrique et se prête ensuite particulièrement bien au développement sélectif des arcs de précipitation.

Technique. Le gel est formé de 0,8 g% de gélose, soigneusement purifiée et d'un tampon véronal de pH 8,6 (soit tampon Michaelis dilué au demi, soit tampon Durrum additionné de lactate de calcium[9]). Le gel est coulé sur des lames de verre 24 h avant l'électrophorèse, de façon à obtenir une très légère déshydratation destinée à faciliter l'absorption de l'échantillon. Les lames de verre utilisées

[1] P. Grabar et C. A. Williams, Biochim. biophys. Acta *10*, 193 (1953).

[2] P. Grabar et. C. A. Williams, Biochim. biophys. Acta *17*, 67 (1955).

[3] J. J. Scheidegger, Int. Arch. Allergy *7*, 103 (1955).

[4] J. Uriel et J. J. Scheidegger, Bull. Soc. Chim. biol. *37*, 165 (1955).

[5] Ch. Wunderly, Exper. *13*, 421 (1957).

[6] R. J. Wieme, *Studies on Agar Gel Electrophoresis* (Arscia, Bruxelles 1959).

[7] J. Uriel, in *Analyse Immunoélectrophorétique* (Ed. P. Grabar et P. Burtin; Masson, Paris 1960), p. 33.

[8] J. Heremans, *Les globulines sériques du système gamma* (Arscia et Masson, Bruxelles et Paris 1960).

[9] W. B. Yeoman, Clin. chim. Acta *4*, 523 (1959).