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CORRESPONDENCE

Differential production of slime by *Staphylococcus saprophyticus* under aerobic and anaerobic conditions

Slime production is an important virulence factor for coagulase-negative staphylococci (CNS) which has been associated with infectious strains, in contrast to colonising or contaminating strains, in enabling them to adhere to smooth surfaces [1]. *Staphylococcus saprophyticus* is a common cause of urinary tract infection in young women [2–4] and it is possible that slime is also a virulence factor for this species [5, 6]. It has been noted that slime production by CNS is media-dependent, and more recently that atmospheric CO_2 content may significantly affect adherence to polymer surfaces [7].

Gatermann and Meyer showed that expression of two major surface proteins of *S. saprophyticus*, one of which is related to adhesion relevant haemagglutinin and the other is *S. saprophyticus* surface-associated protein (Ssp). They also determined the environmental factors that are responsible for the production of these proteins. Anaerobic growth conditions affect the presence of the haemagglutinin protein in the cell wall of *S. saprophyticus* strains. The authors suggested that when these bacteria are grown in an anaerobic atmosphere, haemagglutination titres and fibronectin binding increased, but production of Ssp was suppressed [8].

A total of fourteen clinical isolates were used in this study to determine the role of oxygen in slime production by *S. saprophyticus*. All strains were isolated from specimens of urine from symptomatic female patients. The bacteria were identified by the catalase test, resistance to novobiocin (5-μg disk) and API-Staph (bioMérieux, France). Strains were selected from the collection in the order of receipt to include eight tube adherence weak (Group B) and six tube adherence strong positive (Group A) strains. The ability to produce biofilm was determined for individual colonies of *S. saprophyticus* by a Congo red agar method [9]. *S. saprophyticus* strains 552 (slime positive) and 1132 (slime negative), a gift from Dr Eva Hjelm (University Hospital, Uppsala, Sweden), were used as positive and negative controls for slime production. Tryptic Soy Broth (TSB; Oxoid, CM129) was used because of its recognised ability to promote slime production in CNS. The micro-assay was performed in sterile, polystyrene 96-well flat-bottomed plates. The basis of the assay was that slime-positive cells adhered to polystyrene and cohered to each other, forming a biofilm whose density was measured spectroscopically at a wavelength of 492 nm with a micro-ELISA autoreader, after staining [10].

To determine the role of oxygen in slime production, a culture plate assay was performed under aerobic and anaerobic conditions [11]. To obtain anaerobic conditions, the plates were read after incubation for 24 h at 37°C in anaerobic jars (Gas Pak kit, Oxoid). All assays were performed in duplicate and each microtitration tray included uninoculated medium controls together with positive and negative controls. The paired *t* test was used to compare indices under aerobic and anaerobic conditions. It was observed that strains of both groups (slime production strong positive and slime weak positive) produced significantly more slime under aerobic than anaerobic conditions ($p < 0.005$) (Fig. 1).

We and other authors have reported that slime production by *S. epidermidis* was significantly reduced under anaerobic conditions [12–14]. The results of the present study clearly indicate that slime production by *S. saprophyticus* was also reduced under anaerobic conditions. Thus, it is important to consider the culture environment when assaying for the production of slime in CNS in both clinical and research laboratories.

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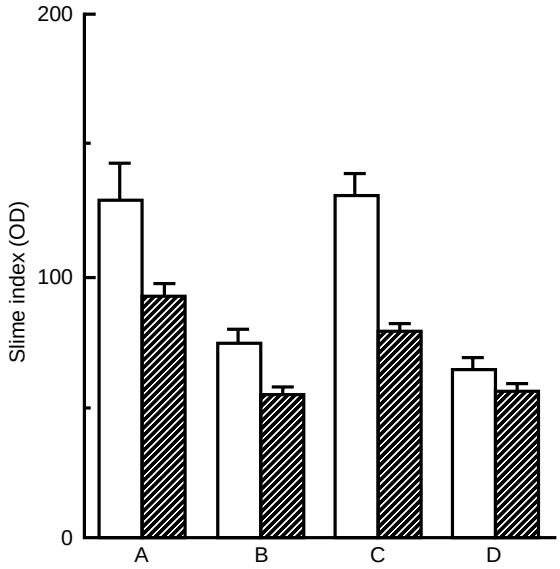

Fig. 1. Aerobic (□) and anaerobic (▨) mean slime production of all strains: group **A** (strong positive), **B** (weak); **C** (552) and **D** (1132) are positive and negative controls, respectively.

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