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followed by removal of the larvae.[10] A broad antibiotic cover is recommended to prevent secondary infections. Our cases illustrate the importance of hygiene and sanitation in tropical countries with high fly population and emphasize the need for correct diagnosis of this obligatory myiasis, which is potentially destructive.

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SOFT TISSUE INFECTIONS WITH ARCANOBACTERIUM HAEMOLYTICUM: REPORT OF THREE CASES

We report here three polymicrobial wound infections associated with *Arcanobacterium haemolyticum* in rural patients aged between 60-65 years. In two patients, one with cellulitis and the other with postoperative wound infection following amputation of the limb, *Arcanobacterium haemolyticum* was isolated repeatedly along with β haemolytic streptococci (BHS). The BHS belonged to Lancefield's group G and group C respectively. In another patient, who was a diabetic with chronic osteomyelitis, *Arcanobacterium haemolyticum* was isolated along with *Proteus vulgaris*. All the three isolates of *Arcanobacterium haemolyticum* isolated by us were uniformly resistant to cotrimoxazole and sensitive to penicillin, erythromycin, clindamycin, ciprofloxacin and gentamicin. Erythromycin alone or combined therapy of penicillin with erythromycin or penicillin with ciprofloxacin was effective in treating these infections.

Key words: *Arcanobacterium haemolyticum, mixed infections, soft tissue infections*

Arcanobacteria are gram positive bacilli which are characteristically catalase negative. As they morphologically resemble *Corynebacterium diphtheriae* they have been grouped under coryneform bacteria.[1,2] To date, three species belonging to *Arcanobacterium* genus: *Arcanobacterium haemolyticum (A.haemolyticum), Arcanobacterium pyogenes* and *Arcanobacterium bernardiae* are thought to be medically relevant.[1]

Arcanobacterium haemolyticum occurs as a commensal on human skin and the mucous membrane of the upper respiratory tract.[2] It is incriminated in causing pharyngitis and soft tissue infections like chronic ulcers and cellulitis. Deep seated infections like sinusitis, endocarditis, meningitis and septicemia due to *A. haemolyticum* have also been reported.[1] The deep seated infections by *Arcanobacterium* spp. are usually monomicrobial whereas the soft tissue infections are polymicrobial, associated with other bacteria.[3] *Arcanobacterium* spp. are also found on the mucous membrane of animals, as in man and cause infections in them.[2]

As arcanobacterial infections have been rarely reported, we report here three polymicrobial wound infections from

which *A. haemolyticum* was isolated along with another co-pathogen.

Case Reports

Case 1

A woman of 64 years, from a village in Bangalore district was admitted to RL Jalappa hospital, Kolar, Karnataka, in June 2004 with the complaints of pain and swelling near lateral malleolus of right foot since two days. There was no history of trauma and the patient was not a diabetic. There was an ulcer over the lateral malleolus from which pus was oozing out. The right leg was swollen and erythematous up to the knee. There was tenderness and local rise of temperature. A provisional diagnosis of cellulitis of right foot was made.

X-ray of the right foot showed diffuse osteoporosis of the fifth metatarsal bone. Haematological study showed leucocytosis and neutrophilia (TC-16,000/cmm,DC-N 87%,L-10%,E-2% and M-1%).

The Gram stained smear of the pus sample from the ulcer showed pus cells along with short filamentous gram positive bacilli with rudimentary branching and gram positive cocci in pairs and chains (Fig. 1a). Culture of the pus sample on sheep blood agar yielded two types of colonies: tiny colonies with wide zones of beta haemolysis and tiny colonies with narrow zones of beta haemolysis. The colonies with wide zone of beta haemolysis were identified as Group G β haemolytic streptococci (GGS) by grouping using latex agglutination and coagglutination tests. The colonies with narrow zone of haemolysis showed wider zones on incubation for 48 hours (Fig. 1b). Gram stained smears of these colonies showed gram positive bacilli with rudimentary branching similar to those seen in the direct smear (Fig. 1b inset). The organism grew on Mueller Hinton agar and brain heart infusion (BHI) agar. It fermented glucose and maltose with the production of acid but no gas and gave a positive reverse CAMP test (Fig. 1c). It did not ferment xylose, sucrose, lactose and mannitol. It was negative for the catalase, urease, gelatin liquefaction and nitrate reduction tests. Based on the above morphological, cultural and biochemical characteristics, the gram positive bacilli were identified as *Arcanobacterium haemolyticum*.[2] A repeat sample yielded the same organisms from the lesion.

Both GGS and *A. haemolyticum*, were sensitive to penicillin, erythromycin, clindamycin, ciprofloxacin and gentamicin. *A. haemolyticum* was resistant to cotrimoxazole and GGS was resistant to tetracycline.

Initially, the patient received cefotaxime intramuscularly 1 g bd for four days with no clinical improvement. After the culture report was available, a below knee POP cast was applied and the patient was administered erythromycin 500 mg tid orally for 10 days, after which she made an uneventful recovery.

Case 2

A man aged 65 years from a village in Kolar district, Karnataka, came to the hospital in December 2004. He was a diabetic under treatment since five years. He had sustained an injury on the right great toe and developed an ulcer on its medial aspect. This was followed by swelling, pain and purulent discharge. When he came to the hospital, there was generalised swelling of distal one third of the right leg and foot. There were two sinuses with foul smelling discharge on the medial and lateral aspects of the right great toe (Fig. 2a). A clinical diagnosis of chronic osteomyelitis of the right great toe was made. X-ray of the right foot showed osteomyelitis of the proximal and distal phalanges of the right great toe (Fig. 2b).

The pus sample collected from the sinus on Gram stain showed many polymorphonuclear cells, gram positive bacilli similar to those seen in case 1 and gram negative bacilli. The pus sample did not show any acid fast bacilli on ZN staining. Culture yielded *A.haemolyticum*, identified as in case 1 and *Proteus vulgaris*. A repeat sample taken from the lesion also yielded both the organisms.

The *A.haemolyticum* from this case had a similar sensitivity pattern as in case 1. *P.vulgaris* was sensitive to ceftriaxone, gentamicin and amikacin. It was resistant to ampicillin, cefazolin, cefuroxime, cotrimoxazole, tetracycline and ciprofloxacin.

The patient was treated with procaine penicillin 4 lakh units intramuscularly and erythromycin 500 mg orally tid, which was started before the culture and sensitivity report was available and continued for six days. Local dressing of the wound was also done with betadine. He improved and there was no discharge from the sinus after eight days. The patient was discharged on request and lost to follow-up.

Case 3

A man aged 60 years from a village near Kolar district, came to our hospital in May 2005 with gangrene of second, third and fourth toes of the right foot. He had sustained an injury 25 days back. On examination lower one third of the right leg was swollen and tender. The gangrenous changes of the skin were evident up to the ankle. He was neither a diabetic nor hypertensive. The haemogram of the patient showed that he was anaemic and there was neutrophilia (Hb-6.8 gm%, TC-23,300 cells/cmm, DC- N 95%,L 5%).

An above knee amputation of the right leg was done. Three days after surgery, the wound showed purulent discharge. The Gram stained smear of the discharge showed few pus cells and gram positive bacilli with rudimentary branching and gram positive cocci in pairs. The pus sample on culture yielded *A. haemolyticum* and Group C Streptococci (GCS). A repeat sample from the patient yielded both *A.haemolyticum* and GCS.

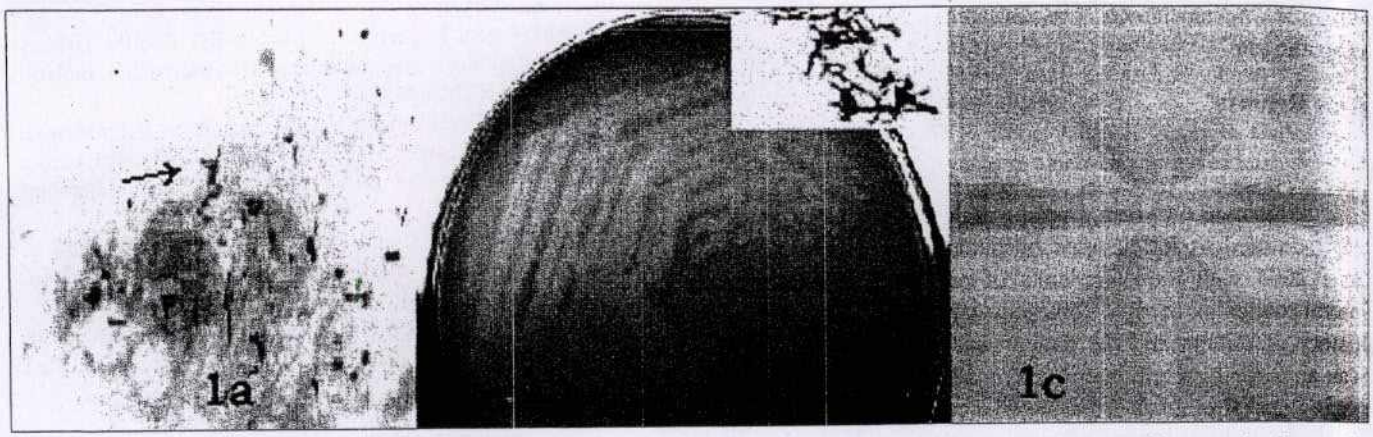

Figure 1: (a) Gram stained smear of the pus sample in case 1, showing branched gram positive bacilli (arrow), gram positive cocci and pus cells (×1000). (b) Case 1: Blood agar plate with beta haemolytic colonies of *A. haemolyticum*. Gram stained smear of the colonies (inset). (c) Case 1: Reverse CAMP test; *A. haemolyticum* is streaked vertically and *S. aureus* is streaked horizontally. Note the zone of inhibition of haemolysis near the streakline of *A. haemolyticum*

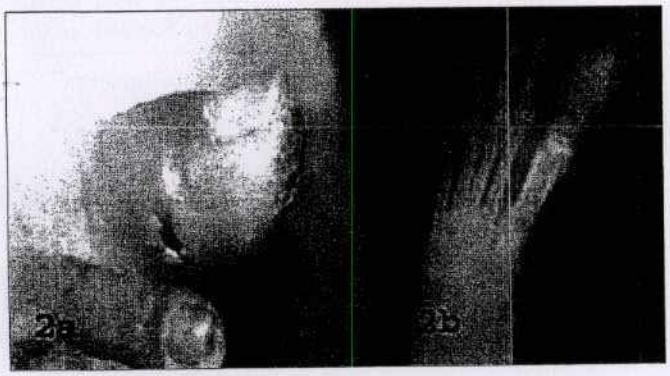

Figure 2: (a) Discharging sinus on the side of the right great toe in case 2. (b) X-ray of the foot shown in Fig. 2a showing osteolysis of the proximal and distal phalanges of the great toe

The antibiogram of *A.haemolyticum* isolated from this patient was similar to the isolates obtained from the earlier two cases. Group C Streptococci was sensitive to penicillin, vancomycin and levofloxacin. It was resistant to bacitracin, erythromycin, cotrimoxazole, chloramphenicol, tetracycline and gentamicin. The patient was administered crystalline penicillin 20 lakh units eighth hourly intravenously and ciprofloxacin 500 mg bd for eleven days. The wound healed satisfactorily.

Discussion

We report here three cases of mixed wound infections caused by *A.haemolyticum* along with a co-pathogen. In the patients reported here, *A.haemolyticum* was associated with β-haemolytic streptococci (BHS) group G and C in cases 1 and 3, respectively and with *P.vulgaris* in case 2. In all the three cases, we could demonstrate the organisms in the direct smear along with pus cells and also repeatedly isolate them from the patients' samples.

Usually the soft tissue infections of *A.haemolyticum* are known to be associated with BHS (Lancefield's groups A, C or G) as in our patients, *Staphylococcus aureus* or *Corynebacterium diphtheriae*.[3] *A.haemolyticum* liberates extracellular enzymes such as phospholipase D, neuraminidase, hemolysin and DNAases which are responsible for the pathogenesis.[1] The co-pathogens are thought to play a synergistic role in causing tissue damage in arcanobacterial mixed infections.[4] *A.haemolyticum* and other potentially pathogenic organisms can exist on skin as bacterial flora. They may invade deeper tissues under favourable local conditions like tissue injury and devitalisation. Systemic conditions like diabetes, chronic alcoholism, immunodeficiency and old age may also predispose to such invasion.[4] Osteomyelitis seen in our second patient exemplifies such invasion and further pathogenesis.

A. haemolyticum strains show *in vitro* sensitivity to β-lactams, vancomycin, macrolides, clindamycin, tetracycline and fluoroquinolones. They are uniformly resistant to cotrimoxazole.[2] Macrolides like erythromycin and azithromycin are considered as the drugs of choice to treat arcanobacterial infections.[1] However, the antibiotic susceptibility pattern of the co-pathogens influence the choice of the antibiotic therapy. In our series, erythromycin was used in the first two cases and penicillin with ciprofloxacin in the third case with good results. Though *Arcanobacterium species* show *in vitro* susceptibility to penicillin, one should bear in mind the possibility of treatment failures reported, probably due to poor penetration of the antibiotic into the bacterial cell.[1]

The review of literature showed that there are very few reports of arcanobacterial infections from India, to date. This could be because *A. haemolyticum* is prone to be missed; it may be wrongly identified as *diphtheroids* based on microscopic morphology or may be passed over as BHS based on colony character on blood agar. Reports from India have documented the following arcanobacterial infections for the first time in literature: arcanobacterial urinary tract infection,[5] association of *A.haemolyticum* with pulmonary

tuberculosis[6] and granulomatous inflammation involving *A.haemolyticum* in osteomyelitis.[7] Our report (case 3) substantiates an earlier report from India[8] that postoperative wound infection following amputation of limb can also occur due to *A.haemolyticum*.

The arcanobacterial infections reported here bring home the point that careful smear examination followed by culture and speciation are important to identify rarer pathogens.

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