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Microbiological features of acute bacterial conjunctivitis in a central Italian area

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SUMMARY

The study aims to identify bacteria causing conjunctivitis in a central Italian area and to analyze chemosusceptibility. From 2005 to 2006, 91 conjunctival swabs were collected from acute conjunctivitis cases and screened for common bacteria and fungi. Susceptibility tests were performed on isolates.
Staphylococcus aureus, *Streptococcus pneumoniae* and *Haemophilus influenzae* amounted to 86.2%. Overall, 100% of strains were susceptible to chloramphenicol and 96.6% to quinolones. Conversely, 20.7% of isolates were tetracycline-resistant and, even if all Gram negative isolates were susceptible to gentamicin, the most frequently isolated pneumococci are constitutively resistant. The study provides support for a rational choice of empiric therapy.

KEY WORDS: Bacterial conjunctivitis, Chemosusceptibility, Therapy

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Acute conjunctivitis is a common presentation in general practice. Most cases of acute conjunctivitis are infectious (Leibowitz, 2000) and bacterial aetiology can account for over 60% of all cases, especially in paediatric patients (Gigliotti, *et al.*, 2000; Block, *et al.*, 1981). The most common aetiologic agents are *Staphylococcus* species, *Streptococcus pneumoniae* and *Haemophilus* species; other rare bacterial pathogens are *Moraxella* spp., *Streptococcus mitis, Streptococcus pyogenes, Corynebacterium diphteriae*, *Neisseria* species and enteric gram-negative rods. Viral and atypical pathogens (e.g. *Chlamydia* spp.) can also be involved, with an almost indistinguishable clinical presentation, especially for general practitioners (Barnes, *et al.*, 2005).

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Although many mild conjunctival infections resolve on their own, antibiotic therapy is used to reduce symptom severity, speed up resolution, eradicate the pathogen and to avoid possible outbreaks (Baum, 1995; Martin, *et al.*, 2003; Hennink, *et al.*, 2006). Topical antibiotics are the gold standard for conjunctivitis: they are easy to administer, achieve a high concentration at the site of action and have few systemic side effects (Baum and Barza, 1983; Leeming, 1999). On the other hand, prescribing antibiotics for minor self limiting illness is generally considered an overmedication, mainly over concern for selective pressure on chemoresistance (Bronzwaer, *et al.*, 2002).
The risk of bacterial resistance is believed to be lower with topical therapy, due to the relatively small number of ocular bacteria and the small concentration of antibiotic that is absorbed systematically (Leeming, 1999). However topical antibiotics were reported as a cause of bacterial resistance in the conjunctiva, cornea and lids (Hwang, 2004) and, moreover, also in extraocular sites (Gaynor, *et al.*, 2005).

Despite these observations, antibiotic prescribing for conjunctivitis has remained high (Everitt and Little, 2002): in clinical practice, physicians nearly always empirically treat acute conjunctivitis and cultures are not normally taken. Lack of a proper laboratory confirmation leads to bacterial agents of ocular infections being unknown in a particular epidemiological setting, no guidance on treatment and overuse of antibiotics in non bacterial illness, with consequent potential selective pressure on resistance.

The study aims to identify bacteria causing conjunctivitis in a central Italian area and to analyze rates of antimicrobial resistance, as a support for clinical management.

From December 2005 to December 2006, 91 conjunctival swabs were collected from patients attending the Ophthalmologic Emergency Room of Siena University (Siena, Tuscany, Central Italy) and having conjunctivitis diagnosed by ophthalmologic specialist clinicians. Informed consent was obtained and the following data were acquired:

1. age;
2. sex;
3. prior empiric antibiotic treatment.

To avoid duplicate isolates, patients with the same culture result at a second visit during 30 days were not included in the study.

Conjunctival swabs were plated on:

1. Columbia agar plus 5% sheep blood;
2. CNA Columbia agar plus 5% sheep blood;
3. Thayer Martin agar;
4. *Haemophilus* selective chocolate agar;
5. Sabouraud dextrose agar plus gentamicin and chloramphenicol (Oxoid Basingstoke, Hampshire, UK).

Pathogen isolation and identification were performed according to standard procedures (Murray, *et al.*, 2003).

Susceptibility tests were performed according to guidelines of Clinical and Laboratory Standards Institutes (CLSI, 2006). Isolates exhibiting resistance to 3 antibiotic classes or more were defined as multiresistant.

Chi-square and, when appropriate, Fisher's Exact tests were used to compare variables. A two-tailed *p* value less than 0.05 was considered statistically significant.

On the total of 91 patients enrolled, 48 (52.7%) were males and 43 (47.3%) females. Twelve subjects (13.2%) were children, five under 2 years of age, seven 2-15 years old; 79 (86.8%) were adults (median 51 years, IQR 35-69.5).

Twenty seven ocular swabs out of 91 (29.7%) were positive for at least one bacterial pathogen, 33.3% in males and 25.6% in females (*p 0.563*): 63% with Gram positive isolation, 30% Gram negative and 7% mixed; no fungal growth was revealed.

Prior empiric antibiotic use was detected in 23 out of 64 culture negative swabs and only 1 out of 27 positive swabs (35.9% *vs* 3.7%, *p 0.001*).

Age group distribution revealed 60% positive swabs among subjects under 2 years old, 42.85% in patients between 2 and 15 years of age and 26.6% in adults (*p 0.207)*.

Staphylococcus aureus, *Streptococcus pneumoniae* and non typable *Haemophilus influenzae* amount to 86.2% of the total isolates (Figure 1); mixed infections *S. aureus* plus *H. influenzae* and *S. aureus* plus *Citrobacter freundii* were revealed in 2 adults patients. Distribution of pathogens by age groups is summarized in Table 1.

Only one strain of *S. aureus* was susceptible to all tested antibiotics, 6 (66.6%) were penicillin-resistant and 2 (22.2%) methicillin-resistant (MRSA). One of the MRSA was a multiresistant strain, showing resistance to all macrolides and lincosamides (MLS phenotype), quinolones and aminoglycosides.

Clinical history of MRSA infected patients was further investigated and risk factors for MRSA acquisition were detected in both of them: one was a previously healthy 7-year-old girl with a probable familial acquisition of a hospital strain

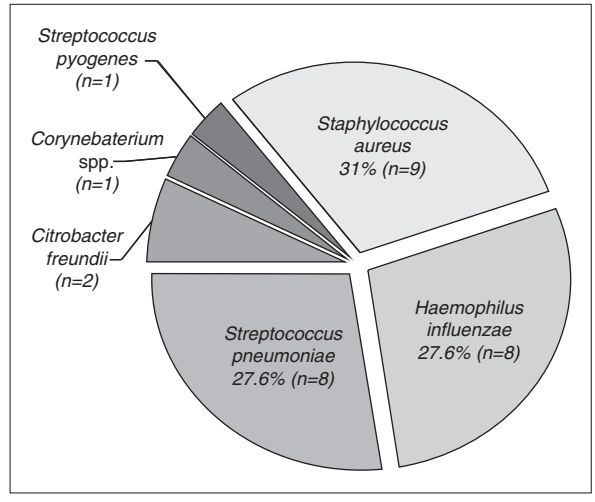

FIGURE 1 - *Isolates from conjunctival swabs.*

TABLE 1 - *Distribution of isolates by age groups.*

	N° (%)		
	<2 years of age	*2-15 years of age*	*≥18 years of age*
S. aureus		1 (33.3%)	6 (28.6%)
H. influenzae	3 (100%)	1 (33.3%)	3 (14%)
S. pneumoniae		1 (33.3%)	7 (33.4%)
S. pyogenes			1 (4.8%)
Corynebacterium spp			1 (4.8%)
C. freundii			1 (4.8%)
Mixed			2 (9.6%)
Total positive swabs (%)	3 (60%)	3 (42.85%)	21 (26.6%)

(mother was a clinical microbiologist), the other one was a 91-year-old woman previously hospitalized for bronchopneumonia and urinary tract infection.

Six (75%) *S. pneumoniae* isolates were susceptible to all tested antibiotics; 2 showed a reduced susceptibility to penicillin (PNSSP, MIC 0.125 mg/l) and one of them was a multiresistant stain (MLS phenotype plus resistance to tetracycline and cotrimoxazole).

Only 1 of the 8 *H. influenzae* strains was a beta lactamase producer; resistance to tetracycline, rifampin and cotrimoxazole was detected respectively in 4 (50%), 2 and 2 (25% each) isolates, one of them revealed a multiresistance to all three antibiotic classes. Remaining bacteria were susceptible to all antibiotics tested except for resistance to ampicillin, amoxicillin plus clavulanic acid, cefaclor and cefoxitin in *C. freundii*. The overall rate of multiresistant isolates was 10.3%.

According to international data (Barnes, *et al.*, 2005), our report confirms *S. aureus*, *H. influenzae* and *S. pneumoniae* as aetiologic agents of nearly 90% of acute bacterial conjunctivitis cases. In particular *H. influenzae* was the only isolate in children under 2 years of age.

Prior empiric antibiotic use can negatively affect *in vitro* bacterial growth: a significant percentage of culture negative patients undertook domiciliary topical antibiotic treatment. Regarding this observation, the rate of bacterial aetiology could be underestimated in our study, even if viral or atypical agents should be considered. We did not judge common routine use searching for viruses in ocular swabs due to expensive and time-wasting techniques: viral infection can be an exclusion diagnosis, based on clinical features and absence of bacterial growth. Equally, we did not test for *Chlamydia trachomatis* that should be considered by clinicians in infants younger than 2-3 weeks and/or in case of no improvement after common antibacterial therapy (Barnes, *et al.*, 2005; Darville, 2006).

Commonly used topical antibiotics seem to be effective against most of isolates: 100% of strains were susceptible to chloramphenicol and 96.6% to quinolones. On the other hand, prescription of tetracycline and aminoglycosides alone should be avoided: 20.7% of all strains were tetracycline-resistant; all Gram negative isolates were susceptible to gentamicin and related antibiotics, but more frequently isolated pneumococcal strains are constitutively resistant.

Regarding other therapeutic options, especially when systemic antibiotic therapy is needed (e.g. conjunctivitis-otitis syndrome), choice of empiric treatment should be based on epidemiological setting knowledge. According to previous epidemiological surveillances (Zanelli, *et al.*, 2002; Cresti, *et al.*, 2003; Montagnani, *et al.*, 2006), our study pointed out that: (i) the rate of PNSSP seems to be significant, (ii) MRSA and beta lactamase producer *H. influenzae* isolation is possible, though it might be quite rare.

When clinical history (e.g. previous hospitalization or antibiotic use, health care workers) suggests a multiresistant bacterium, conjunctival swabs should be mandatory before empiric therapy, even if it is not usually a common routine step in conjunctivitis management.
In conclusion, this study provides support for a rational empiric therapy, but whenever possible a prior swab seems to be useful to avoid over-medicalisation, guide therapy in case of therapeutic failure and constantly monitor the local setting.

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