Low rate of co-infection in complicated infectious mononucleosis

Christian Sander Danstrup & Tejs Ehlers Klug

ABSTRACT

INTRODUCTION: It remains unclarified if bacterial co-infection is common in patients with infectious mononucleosis (IM) and acute tonsillitis and/or peritonsillar abscess (PTA). Recent studies suggest that *Fusobacterium necrophorum* is a prevalent pathogen in acute tonsillitis and PTA. We hypothesised that this anaerobe may play a significant role for the aggravated infection and the development of PTA among teenagers and young adults with IM.

METHODS: All patients with IM and clinical findings of acute tonsillitis or PTA admitted to our department in the 2001-2015 period were included in the study.

RESULTS: In total, 257 patients with IM and acute tonsillitis (n = 220) or PTA (n = 37) were included. Positive bacterial cultures were obtained in 28% of patients with AT and in 50% of PTA patients. The most prevalent bacterial findings were Group C/G streptococci (14%) among patients with acute tonsillitis and *Staphylococcus aureus* (22%) in PTA patients. *F. necrophorum* was recovered in 9% and 2% of patients with acute tonsillitis and PTA, respectively.

CONCLUSIONS: We were unable to substantiate a prevalent role for *F. necrophorum* in patients with IM and acute tonsillitis/PTA. S. aureus may play a role in PTA development in IM patients. The majority of our findings did not support the use of antibiotics in patients with IM, even in this selected group of patients with severe symptoms and a high risk of PTA.

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Infectious mononucleosis (IM) is a common viral infection caused by Epstein-Barr virus (EBV). In early childhood, symptoms may resemble those of other upper respiratory tract virus, whereas adolescents and young adults often present with fever, acute tonsillitis (AT) and cervical lymphadenopathy [1]. In severe cases, hyperplasia of the lymphoid tissues in Waldeyer's tonsillar ring may cause upper airway obstruction (**Figure 1**) [2]. Treatment of IM is commonly symptomatic and does not require admission. However, patients suspected of having suppurative complications or severe AT with respiratory distress or dehydration are often admitted to ear-, nose- and throat (ENT) departments.

A wide range of other acute complications have been described, including peritonsillar abscess (PTA), haemolytic anaemia, thrombocytopenia, neutropenia and hepatitis [2, 3].

It is unclear if bacterial co-infection is common in EBV-positive AT. Stenfors and colleagues found weakened anti-bacterial defences on the tonsillar mucosa membrane, increased bacterial penetration into the epithelial cells and massive bacterial colonisation on the tonsils [4-6]. However, the significance of this colonisation remains largely unexplored, the pathogenic bacteria are unclarified, and it is controversial if patients with EBV-positive AT profit from antibiotic therapy. Recent studies suggest that *Fusobacterium necrophorum* is a prevalent pathogen in AT and PTA [7, 8]. This anaerobic bacterium is especially prevalent in tonsil-derived infections among patients aged 15 to 30 years [9]. These life stages coincide with clinically severe EBV-positive AT and PTA, and we hypothesised that *F. necrophorum* may play a significant role for the aggravated infection and the relatively frequent development of PTA among teenagers and young adults with IM.

The purpose of this study was to describe the clinical, biochemical and bacterial findings in patients admitted to an ENT department with severe IM, with a special attention to the presence of *F. necrophorum*. Furthermore, we aimed to describe our management and evaluate if patients were likely to benefit from antibiotic treatment using the bacterial findings and outcome in patients with or without antimicrobial therapy prior to, during and after admission.

METHODS

Included in the study were all patients admitted to the Department of Otorhinolaryngology, Aarhus University Hospital, Denmark, in the 2001-2015 period with IM and clinical findings of AT or PTA. The hospital has a catchment area of 650,000 patients.

The diagnosis of IM was based on the finding of atypical lymphocytosis (lymphocyte count > 3.50 × 10^9/l) and/or a positive mononuclear spot (Monospot) test at the time of admission. In this clinical study, we

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Department of Otorhinolaryngology, Head and Neck Surgery, Aarhus University Hospital, Denmark

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were unable to use the comprehensive gold standard criteria for IM [10]. However, we included patients who were very likely to have EBV-positive IM and who were treated accordingly. The addition of lymphocytosis (to the monospot test) as an inclusion criterion was chosen to counteract the risk of missing false-negative IM patients due to the monospot test's decreased sensitivity among children [11] and within the first week of illness in general [12].

Cultures were made from tonsillar surface swabs or pus aspirates or swabs. Culturing and identification of bacteria from these specimens were performed as part of the routine diagnostic procedures. Briefly, blood agar, chocolate agar, anaerobic plates and thioglycolate broth (SSI Diagnostic, Hilleröd, Denmark) were used to culture the specimens. The cultures were incubated at 35 °C in a CO_2-enriched atmosphere and anaerobically for up to three days. Speciation for microorganisms was performed by standard methods [13]. Light to moderate growth of viridans group streptococci, *Neisseria* species, *Lactobacillus* species, coagulase-negative staphylococci, *Prevotella* species and Fusobacterium non-necrophorum alone or in mixture were reported as "mixed tonsillar flora".

The study was approved by the Danish Data Protection Agency. In accordance with Danish law, the study was not reported to the local ethical committee. Statistical analyses were performed using Fisher's exact test, Students t-test and the Kruskal-Wallis test. Statistical significance was defined as $p < 0.05$.

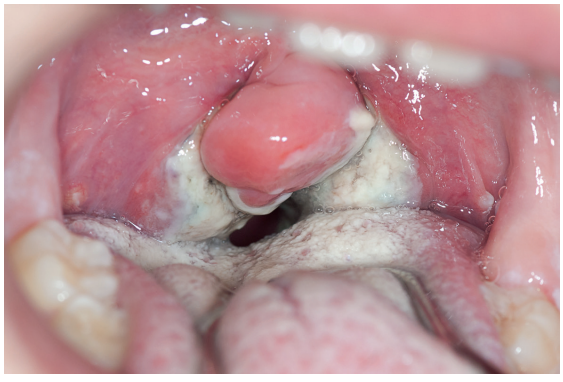
Trial registration: The Danish Data protection Agency approved the project.

RESULTS

In total, 257 patients with IM and either AT (n = 220) or PTA (n = 37) were included in the study. Increased lymphocyte count was found in 201 (78%) patients and the Monospot was positive in 217 of 230 tested patients (94%). Males (58%) were significantly more prevalent than females (42%) ($p < 0.001$, Fisher's exact test) (**Table 1**). Males (median 18 years) were significantly older than females (median 16 years) ($p = 0.010$, Kruskal-Wallis test). The absolute neutrophil count was significantly higher in patients with PTA (mean 6.7×10^9/l) than in patients with AT (mean 5.3×10^9/l) ($p = 0.016$, Student's t-test). No statistically significant differences between patients with AT and PTA were found for age, duration of symptoms, duration of admission or C-reactive protein levels (Table 1). The mean annual incidence of IM and AT or PTA that required admission was 2.8 cases/100,000 population.

FIGURE 1

Patient with infectious mononucleosis. Hypertrophic tonsils covered by "whitewash" exudate. Oedema of the uvula and oropharyngeal hyperaemia. Photo: Copyright Tejs Klug.



Bacterial findings

Bacterial cultures were performed from tonsillar surface swabs in 57 patients with AT and from pus aspirates or swabs in 32 patients with PTA (**Table 2**). Group C/G streptococci (14%) were the most prevalent finding in AT patients, whereas *Staphylococcus aureus* (22%) was predominant in PTA patients. *F. necrophorum* was recovered from 2% (1/57) of AT patients and 9% (3/32) of patients with PTA.

Management

Prior to admission, antibiotics were prescribed to 45% of patients. The drugs of choice were phenoxymethylpenicillin, macrolides, amoxicillin and other antibiotics in 86%, 5%, 5% and 3% of cases, respectively.

Significantly fewer PTA patients (27%) than AT patients (48%) received antibiotics prior to admission ($p = 0.020$, Fisher's exact test). During hospitalisation, 65% of the patients were treated with antibiotics. The drugs of choice were benzyl-penicillin (87%), phenoxymethyl-penicillin (6%), cefuroxime (4%), macrolides (1%), amoxicillin (1%) and metronidazole (1%). After discharge, 37% of patients were prescribed antibiotics (phenoxymethyl-penicillin (91%), macrolides (4%), amoxicillin (2%), benzyl-penicillin (1%) and metronidazole (1%). The standard dose of benzyl-penicillin was two million IU three times daily.

Patients with AT who were treated with antibiotics during hospitalisation and upon discharge had significantly higher C-reactive protein levels (mean 69.0 mg/l and 80.2 mg/l, respectively) than patients who were not prescribed antibiotics during hospitalisation and upon discharge (mean 53.5 mg/l and 54.0 mg/l, respectively) ($p = 0.031$ and $p < 0.001$, Student's t-test, respectively).

Acute bilateral tonsillectomy was performed in 53 (21%) patients. The indications for tonsillectomy were

TABLE 1

Demographic, antimicrobial therapy and biochemical data in 257 patients admitted with infectious mononucleosis, stratified by type of infection and surgical intervention.

	Acute tonsillitis				Peritonsillar abscess				
	total (n = 220)	tonsillectomy (n = 20)	no surgery (n = 200)	p-value	total (n = 37)	tonsillectomy (n = 33)	incision and drainage (n = 4)	p-value	p-value[a]
Males, %	59	60	58	0.35[c]	54	55	50	1.00[c]	0.6 0[c]
Age, median (range), yrs	17 (2-46)	17 (5-23)	17 (2-46)	0.44[b]	17 (4-36)	17 (4-32)	26 (16-36)	0.08[b]	0.11[b]
Duration of symptoms, median (range), days	6 (1-31)	6 (1-21)	6 (1-31)	0.69[b]	7 (2-14)	7 (2-14)	6 (2-8)	0.56[c]	0.75[b]
Antibiotics, %									
Prior to admission	48	65	46	0.16[c]	37	24	50	0.29[c]	0.020[c]
During hospitalisation	62	80	60	0.10[c]	84	82	100	1.00[c]	0.014[c]
Upon discharge	37	40	37	0.812[c]	32	24	100	0.007[c]	0.71[c]
Duration of hospitalisation, median (range), days	2 (0-13)	3 (2-7)	2 (0-13)	< 0.001[b]	3 (2-7)	3 (2-7)	3 (2-3)	0.73[b]	0.39[b]
C-reactive protein, mean (± SD), mg/l	65 (± 54)	68 (± 50)	65 (± 55)	0.21[d]	80 (± 56)	74 (± 52)	122 (± 85)	0.11[d]	0.10[d]
Absolute neutrophil count, mean (± SD), × 10^9/l	5.3 (± 2.6)	4.8 (± 2.1)	5.3 (± 2.6)	0.50[d]	6.7 (± 4.7)	6.7 (± 4.7)	7.3 (± 4.4)	0.80[d]	0.016[d]
Lymphocyte count, mean (± SD), × 10^9/l	6.1 (± 3.2)	6.0 (± 2.9)	6.1 (± 3.2)	0.82[d]	5.0 (± 2.6)	5.2 (± 2.6)	3.4 (± 1.4)	0.20[d]	0.037[d]

SD = standard deviation.
a) Acute tonsillis vs peritonsillar abscess.
b) Kruskal-Wallis test.
c) Fisher's exact test.
d) Student's t-test.

suspected PTA in 34 cases (PTA was found in 28 cases) and tonsillar hypertrophy with upper airway obstruction in 19 cases (PTA was found in five cases). Four patients with PTA underwent incision and drainage in local anaesthesia. Nine (25%) PTA patients had bilateral abscesses.

Complications

At the time of admission, 22 (10%) patients with AT and three (8%) patients with PTA had respiratory distress. Twelve (5%) patients were readmitted after discharge because of pain or general deterioration (n = 7), post tonsillectomy haemorrhage (n = 3) or PTA development (n = 2).

DISCUSSION

In accordance with the literature, the majority of patients admitted with IM were teenagers and young adults [14]. We found a statistically significant male preponderance and that females were significantly younger than males. These findings seem novel for IM patients. However, in a previous study from our institution, patients with PTA had similar age and gender associations regardless of aetiology [9]. A significant proportion (14%) of our patients admitted with IM had PTA, which seems higher than previously reported [15]. Furthermore, PTA was found in five of 19 patients who were tonsillectomised due to tonsillar hypertrophy and even more patients may have had minor, undiagnosed PTAs. Also stressing the high risk of PTA development in severe IM, bilateral PTA was found in 25% of patients, which is significantly higher than described in the literature of non-IM PTA (average 4.1%) [16].

Our hypothesis that *F. necrophorum* is prevalent in patients with IM and AT/PTA was not substantiated. In fact, *F. necrophorum* was only recovered from 9% of

TABLE 2

Bacterial findings in 89 cultures from 257 patients with infectious mononucleosis.

	Acute tonsillitis	Peritonsillar abscess
Cultures, n (%)		
Mixed tonsillar flora[a]	41 (72)	16 (50)
Positive cultures	16 (28)	16 (50)
Total	57	32
Isolates, n	17	20
Organisms, n (% of cultures)		
Fusobacterium necrophorum	1 (2)	3 (9)
Group A streptococci	2 (4)	3 (9)
Group C or G streptococci	8 (14)	3 (9)
Staphylococcus aureus	2 (4)	7 (22)
Haemophilus influenzae	1 (2)	2 (6)
Other aerobes	3 (5)	1 (3)
Other anaerobes	-	1 (3)

a) Growth of viridans streptococci, *Neisseria* species, *Lactobacillus* species, coagulase-negative staphylococci, *Prevotella* species, and *Fusobacterium* non-necrophorum alone or in mixture.

PTA pus cultures and 2% of tonsillar swabs from AT patients, which is less than in PTA and AT patients without IM [16].

In a previous study of tonsillar core bacteria, *S. aureus* was recovered significantly less frequently and in significantly lighter growth in patients with PTA than in non-infected controls [7]. In the present study, *S. aureus* was found in 22% of PTA pus cultures making it the most prevalent potential pathogen. Furthermore, five of the seven isolates were recovered in pure culture. We were unable to clarify if this frequent finding of *S. aureus* signifies a less altered normal flora or pathogenic significance. Group A streptococci were obtained in only 9% of cases, which is less than in the literature of non-IM PTA [16]. Hence, the pathogens associated with PTA development in IM patients seems different from those in non-IM PTA, and one may speculate if bacteria are involved in the pathogenesis of PTA in all cases among EBV-positive patients? Some findings in the present study point towards bacterial co-infection in patients with PTA compared with AT: the absolute neutrophil count was higher in patients with PTA than in patients with AT ($p = 0.016$), there was a trend towards more frequent positive cultures in patients with PTA than in patients with AT ($p = 0.065$), and fewer PTA patients than AT patients had received antibiotics prior to admission ($p = 0.020$). On the contrary, the absolute neutrophil counts were within the normal range ($2.0\text{-}7.0 \times 10^9$/l) in 68% of PTA patients and the duration of symptoms was the same for patients with PTA and AT. Only a limited number of studies have been conducted studying the tonsillar flora in patients with IM and AT [17, 18]. Based on intensive cultures at the time of admission and 60 days later, Brook and de Leyva concluded that *S. aureus, F. nucleatum* and *Prevotella intermedia* were potential co-pathogens.

It is noteworthy that antibiotics were prescribed to almost half of the patients prior to admission. We were unable to determine the proportion of patients who were diagnosed with IM prior to admission. However, based on the infrequent finding of Group A streptococci, it seems unlikely that antibiotics were prescribed secondarily to a positive streptococcal antigen detection test in more than a few cases. Even after the diagnosis, a significant proportion of patients were treated with antibiotics.

Only few studies have examined the effects of antibiotics in IM patients. Hedström et al found that oral metronidazole reduced the duration of symptoms and fever [19]. However, in a randomised double-blinded clinical trial, metronidazole had no effect on the duration or intensity of symptoms [20].

The majority of our findings do not point to beneficial effects of antibiotics in patients with IM and AT. Firstly, only 28% of bacterial cultures were positive for potential pathogens and the findings resembled those of non-infected tonsils [7]. Secondly, similar proportions of patients with or without respiratory distress or other complications received antibiotics prior to admission. Thirdly, similar proportions of patients, who were re-admitted because of PTA development or progression of symptoms, were treated with antibiotics during hospitalisation and after discharge. Lastly, no difference in the length of hospitalisation was found between patients treated with antibiotics prior to and during admission compared with non-treated patients. On the contrary, significantly fewer patients with PTA (27%) than with AT (48%) were treated with antibiotics prior to admission, which may suggest that antibiotics reduce the risk of PTA development. All these analyses carry the multiple potential biases of a non-randomised study and should therefore be interpreted with great caution. Nevertheless, the beneficial effects of antibiotics in IM patients, if any, seem very modest.

One strength of the present study is the fact the majority of data were collected prospectively during the study period. In addition, the cohort was relatively large and consisted of consecutive patients.

Our study also has several significant limitations. The observatory study design prevents solid conclusions on the effects of antibiotic treatment of patients with IM due to multiple potential biases and confounders. In addition, we were unable to obtain information on the duration of and compliance with antibiotic treatment prior to admission and after discharge, which may lead to underestimation of the effects of antibiotics. Some patients who had lymphocytosis – but tested negative by the Monospot test – may not have had EBV infection. However, the demographic, clinical and biochemical characteristics were similar for patients with a negative test or a positive test. Lastly, the cohort of patients with IM is highly selected as we only included admitted patients and our findings should be interpreted in this context.

CONCLUSIONS

Our hypothesis that *F. necrophorum* is prevalent in patients with IM and AT/PTA was not substantiated. The significant pathogens in IM-positive PTA remain unidentified, and we question if bacteria are involved in the pathogenesis of all PTA cases among patients with IM. The majority of our findings did not support the use of antibiotics in patients with IM, even in this selected group of patients with severe symptoms and a high risk of PTA.

CORRESPONDENCE: *Christian Sander Danstrup.*
E-mail: Christian.danstrup@gmail.com

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