Oral Microflora: A Comparative Study in HIV and Normal Patients

Mahesh Chandra Hegde · Abhijit Kumar ·
Gopalkrishna Bhat · Suja Sreedharan

Received: 9 July 2011 / Accepted: 15 November 2011 / Published online: 30 November 2011
© Association of Otolaryngologists of India 2011

Abstract The study was designed to compare the oral microbiota in normal and HIV-infected individuals. The study tries to establish a significant shift in oral microflora in HIV-infected patients. Antibiotic sensitivity testing was performed to establish any rise in resistance against the antibiotics. It was a two and half year prospective study conducted in a tertiary care centre. The study group consisted of eighty subjects divided into two groups of control and HIV. The age range for this group was 9–75 years. The mean age in this group was 39.7 years. The male:female ratio was 2.75:1. Tuberculosis was the most common opportunistic infection in patients with HIV infection. The most common commensal micro organism isolated was the Viridans streptococci in 60% followed by *Streptococcus pneumoniae* in 23.33%. HIV Group: The most common commensal micro organism isolated was the Viridans streptococci in 42%; this was followed by the *Micrococci* spp. in 22% cases. *S. pneumoniae* was isolated in 6% of cases. The colony count for Viridans streptococci showed a heavy growth in 55.56% of cases in controls whereas the same in HIV group was 62.5%. *Micrococcus* spp. was isolated from 11 subjects in HIV group while it was not isolated from the controls. 50% subjects in the HIV group showed a heavy growth of *Klebsiella* spp. whereas controls showed only moderate and scanty growth. In patients with CD4+ T cell count less than 50 cells/ μ l we found a heavy colonization of the oral cavity with *Micrococcus* spp.,

Acinetobacter and *Klebsiella* spp. Viridans streptococcus was not isolated in any of the patients with CD4+ T cell count less than 50 cells/ μ l. As CD4+ T cells counts improved to 51–100 cells/ μ l Viridans streptococcus colonies returned and 37.5% patients showed a heavy growth. *Micrococcus* spp. colonies were isolated till the CD4+ T cells improved up to 300 cells/ μ l. At counts > 300 cells/ μ l the oral microbiota became comparable to that of the controls. Many of the opportunistic infections in HIV are caused by commensal bacteria which are otherwise harmless in a normal individual. Our study is unique in that such a study of the oral commensals in HIV patients has never been reported. We found an increased colonization of the oral cavity by *Micrococcus* spp. which is a normal commensal of the skin.

Keywords HIV · Normal · Oral microflora · *Micrococcus*

Introduction

Acquired Immunodeficiency Syndrome (AIDS) has killed more than 25 million people since it was first recognized in 1981, making it one of the most destructive epidemics in recorded history [1]. India has a population of one billion, around half of whom are adults in the sexually active age group. The first AIDS case in India was detected in 1986, and since then HIV infection has been reported in all states and union territories. The Indian National Aids Control Organization (NACO) estimates that 5.134 million people were living with HIV in 2004. The adult prevalence rate of HIV in India is 0.9% [2].

Acute infection is the leading cause of morbidity and mortality in immunosuppressed patients with HIV infection. In neutropenic patients, it's a well known fact that

M. C. Hegde (✉) · G. Bhat · S. Sreedharan
Department of Otolaryngology, Kasturba Medical College
Hospital, Attavara, Mangalore 575001, India
e-mail: drak30@gmail.com

A. Kumar
Vinayaka Missions Kripananda Variyar Medical College, Salem,
Tamil Nadu, India

acute exacerbations of pre existent periodontal disease can result in systemic sequelae [3]. In HIV infection, like all viral infection it is believed that the cell-mediated immunity plays a very important role in host defense. Acute infections of the upper respiratory tract are prevalent in all stages of HIV infection. The most severe cases tend to occur in patients with lower CD4+ T cell counts. HIV-infected individuals are predisposed to infection with bacteria; in particular *Streptococcus pneumoniae* and non-typhi *Salmonella* [4]. Other studies have found an increased incidence of infection with *Haemophilus influenza* and *Pneumococci* which are capsulated organisms [5]. Schmidt et al. [6] in his study found that 5.0% of the HIV-infected patients had colonization with different species of *Enterobacteriaceae*.

Several studies have established the indiscriminate use of antimicrobials may cause a shift of oropharyngeal microbiota and favor colonization with gram-negative rods [7, 8]. Such a shift is well established in patients with leukemia [9], but there are few published accounts highlighting such a shift in HIV-infected patients.

The present study was designed to compare the oral microbiota in normal and HIV-infected individuals. The study tries to establish a significant shift in oral microflora in HIV-infected patients. Antibiotic sensitivity testing was performed to establish any rise in resistance against the antibiotics.

Patients and Methods

This study was conducted at the Kasturba Medical College, Mangalore, Karnataka, India. It was a two and half year prospective study. The patients attending the outpatient department (OPD) of the Department of Otorhinolaryngology at the Kasturba Medical College and its associated hospitals were included in the study.

The study group consisted of eighty subjects divided into two groups of control and HIV. All the subjects were further subdivided based on age, sex, and socioeconomic status and data analysis was done using SPSS 14.0.

Exclusion Criteria

All subjects in the study group should have no evidence of

1. Upper respiratory tract infection,
2. Local lesions like oral ulcers, follicles in the tonsils, tumours, leukoplakia, erythroplakia and candidiasis.
3. Controls without HIV infection but with immunodeficiency states, both genetic and acquired like chronic liver disease, diabetes, transplant recipients and cancer patients on treatment.

4. Patients with HIV infection on antiretroviral drugs.
5. Patients who are receiving antibiotics or those who have received antibiotics in the last 2 weeks.

HIV infection was diagnosed by HIV ELISA and confirmation was done by western blot [10]. Once included in the study all the subjects were asked a detailed clinical history, followed by a detailed clinical examination. Routine investigations like complete blood count, CD4 count and urine routine were performed.

Specimen Collection [9]

We used 0.85 to 0.9% normal saline mouth gargles or oral washings for collecting specimens. Subjects were asked to gargle for 1 min and then the contents are collected in a sterile container. Early morning samples were collected and samples were transported to the lab within 30 min.

Microbiological Techniques

Once in the laboratory the washings were inoculated in 5% sheep blood agar for aerobic culture using standard calibrated loop with an internal diameter of 4 mm. The inoculated plates were incubated for 24–48 h at 37°C. Colony count/ml was determined by multiplying the number of colonies by 100.

Identification of Bacteria [11]

The bacteria were identified by colony morphology, gram staining, test for motility, catalase test, oxidase test and other biochemical tests. *Staphylococcus aureus* was identified by colony morphology, pigment production, gram staining, and catalase test, fermentation of mannitol and coagulase test.

Antibiotic Sensitivity Tests

Antibiotic sensitivity for the pathogens was done by the Kirby Bauer disc diffusion method (Bauer AW, Kirby WMM, 1966) using commercially available antibiotic discs (Hi-Media, Bombay). For non-fastidious bacteria, Mueller–Hinton agar and for fastidious bacteria, Mueller–Hinton agars supplemented with 5% sheep blood were used. The antibiotic discs used were: ampicillin (10 µg), amoxicillin (30 µg), amikacin (30 µg), chloramphenicol (30 µg), cefotaxime (30 µg), ciprofloxacin (5 µg), ceftriaxone (30 µg), co-trimoxazole (25 µg), cephazoline (30 µg), erythromycin (15 µg), gentamicin (10 µg), oxacillin (1 µg), penicillin G (10 µg), piperacillin (100 µg), tetracycline (30 µg), and tobramycin (10 µg). The plates were incubated at 37°C for

24 h and the results were interpreted based on the guidelines given by the National Committee for Clinical Laboratory Standards (NCCLS, 1990).

Results and Observations

Eighty patients were included in the study. The patients were subdivided into thirty normal (Control) and fifty HIV-infected patients (HIV).

Age and Sex Incidence

Control

The age range for this group was 9–75 years. The mean age in this group was 39.7 years (SD 15.8, Variance 249).

The male:female ratio was 2.75:1 (Table 1).

HIV

The age range was 24–66 years with a mean age of 38.4 years (SD 8.87, Variance 78.67).

The male:female ratio was 5.25:1.

In Control group the most common age group affected was the 31–40 years, followed by the 21–30 years age group with 33.33 and 16.67%, respectively. In the HIV group the most common age group affected was the 31–40 years with 54% of the subjects in this group (Table 1).

CD4+ T Cell Count

32% of subjects in HIV group had a CD4+ T cell count in the range of 51–100 cells/ μ l (Table 2). The mean CD4 value in HIV group was 179 (range 9–500). Only 2% of the HIV patients had CD4 counts more than 500.

Table 1 Age and sex incidence

Age groups	Control		HIV		Total
	Male	Female	Male	Female	
<20	03	01	00	00	04
21–30	03	02	06	03	14
31–40	06	04	24	03	37
41–50	04	00	10	01	15
51–60	04	00	00	01	05
>60	02	01	02	00	05
Total	22	08	42	08	80

Table 2 CD4 counts in both groups

CD4 count	Control	HIV	Total	% (HIV)
0–50	00	09	09	18
51–100	00	16	16	32
101–200	00	12	12	24
201–300	00	03	03	06
301–400	00	06	06	12
401–500	00	03	03	06
>500	30	01	31	02
Total	30	50	80	100

Table 3 Organisms isolated in control group

Organisms isolated	Number	%
Viridans streptococci	18	60
<i>Streptococcus pneumoniae</i>	07	23.33
<i>Acinetobacter</i>	04	13.33
<i>Moraxella catarrhalis</i>	04	13.33
<i>Klebsiella</i> spp.	02	6.67

Organisms Isolated

Controls

The most common commensal micro organism isolated was the Viridans streptococci in 60% followed by *S. pneumoniae* in 23.33%. The other commensals isolated are shown in Table 3.

HIV Group

The most common commensal micro organism isolated was the Viridans streptococci in 42%; this was followed by the *Micrococci* spp. in 22% cases. *S. pneumoniae* was isolated in 6% of cases (Table 4). The most common isolate in both the groups were Viridans streptococci. *Micrococci* spp was isolated in 22% patients in HIV group compared to 3% in normal group ($P = 0.005$, $SD = 6.22$).

Colony Count

The colony count for Viridans streptococci showed a heavy growth in 55.56% of cases in controls whereas the same in HIV group was 62.5% (Table 5). *Micrococcus* spp. was isolated from 11 subjects in HIV group while it was isolated in one control patient. A heavy growth of *Acinetobacter* was found in 75 and 50% subjects in HIV group and controls, respectively. 50% subjects in the HIV group showed a heavy growth of *Klebsiella* spp. whereas controls showed only moderate and scanty growth. We found a

Table 4 Organisms isolated in HIV group

Organisms isolated	Number	%
Viridans streptococci	21	42
<i>Micrococcus</i> spp.	11	22
<i>Acinetobacter</i>	08	16
<i>Klebsiella</i> spp.	08	16
<i>Streptococcus pneumoniae</i>	03	06
<i>Bacillus</i>	02	04
<i>Moraxella catarrhalis</i>	01	02

heavy growth of *S. pneumoniae* in HIV group (66.67%) whereas moderate growth in control group (71%).

Correlation Between CD4+ T Cell Count and Microbiota HIV Group

In patients with CD4+ T cell count less than 50 cells/ μ l we found a heavy colonization of the oral cavity with *Micrococcus* spp., *Acinetobacter* and *Klebsiella* spp. (Table 6). Viridans streptococcus was not isolated in any of the patients with CD4+ T cell count less than 50 cells/ μ l. As CD4+ T cells counts improved to 51–100 cells/ μ l Viridans streptococcus colonies returned and 37.5% patients showed a heavy growth. *Micrococcus* spp. colonies were isolated till the CD4+ T cells improved up to 300 cells/ μ l. At counts > 300 cells/ μ l the oral microbiota became comparable to that of the controls.

Antibiotic Sensitivity Testing

The antibiotic sensitivity pattern in both the groups showed sensitivity to the most commonly used antibiotics. There was no evidence of any increase in antibiotic resistance among organisms even when the CD4+ T cell count was less than 50 cells/ μ l.

Table 5 Colony count and organisms

Organism	Groups	Heavy	Moderate	Scanty
Viridans streptococci	Control	10	08	00
	HIV	15	06	03
<i>Micrococcus</i> spp.	Control	00	01	00
	HIV	11	00	00
<i>Acinetobacter</i>	Control	02	00	00
	HIV	06	00	00
<i>Klebsiella</i> spp.	Control	00	01	00
	HIV	04	03	01
<i>Streptococcus pneumoniae</i>	Control	02	05	00
	HIV	02	00	01

Heavy >10⁵ CFU/ml, moderate 10³–10⁴ CFU/ml, scanty <10³ CFU/ml

Table 6 Correlation between CD4+ T cell count and microbiota in HIV group

CD4 count (cells/ μ l)	Organism	Number	Colony count (CFU/ml)
<50	AB	2	>10 ⁵ [5]
	MIC	2	>10 ⁵
	KL	2	>10 ⁵
	VS	2	10 ³ –10 ⁴
	SE	1	10 ³ –10 ⁴
	VS	6	>10 ⁵
	MIC	4	>10 ⁵
51–100	KL	1	<10 ³
	EN	1	10 ³ –10 ⁴
	MX	1	>10 ⁵
	PC	1	<10 ³
	SA	1	>10 ⁵
	AB	1	>10 ⁵
	VS	3	>10 ⁵
	MIC	3	>10 ⁵
	KL	2	>10 ⁵
	N	1	>10 ⁵
101–200	AB	1	>10 ⁵
	EN	1	>10 ⁵
	PC	1	>10 ⁵
	SE	1	>10 ⁵
	VS	2	>10 ⁵
	MIC	1	>10 ⁵
	KL	1	>10 ⁵
	AB	1	>10 ⁵
	VS	4	>10 ⁵
	KL	2	>10 ⁵
201–300	SA	1	>10 ⁵
	AB	1	>10 ⁵
	VS	2	>10 ⁵
	BAC	1	>10 ⁵
	PC	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
301–400	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
401–500	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
>500	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵
	VS	1	>10 ⁵

AB *Acinetobacter*, VS *Viridans streptococci*, PC *Streptococcus pneumoniae*, BAC *Bacillus* spp., SA *Staphylococcus aureus*, KL *Klebsiella*, MIC *Micrococcus*, SE *Staphylococcus epidermidis*, EN *Enterobacteriaceae*, N *Neisseria* spp., MX *Moraxella*

Discussion

HIV-infected patients especially children have an exceptional vulnerability to invasive bacterial infections that is much greater than that seen in immunocompetent persons. Numerous defects in the immunologic system are responsible for the increased vulnerability of the HIV-infected to serious bacterial illness. These include defects in the cell-mediated and the humoral arms of the immune system; phagocytic abnormalities including decreases in neutrophil number, multiple defects in neutrophil function and

impairment in macrophage and monocyte function [12]; functional asplenia and defects in three components of complement [13]. These defects become more severe as the HIV disease progresses. Other factors that increase susceptibility to infection in the HIV-infected include frequent use of broad-spectrum antibiotics, frequent hospitalizations, use of indwelling intravascular catheters that disrupt the integrity of skin, malnutrition, micronutrient deficiencies, and lack of adequate medical care. The results of these defects are: increased susceptibility to infection with encapsulated bacteria beyond age of 2 years, [14] increased nasopharyngeal colonization rates for *S. pneumoniae* and *H. influenzae*, [15] recurrent infections with the same bacterial species, increased susceptibility to infections with bacteria unusual in immunocompetent hosts and, increased morbidity and mortality.

Age and Sex Incidence

Sarkar K et al. [16]. in their study of female sex workers found HIV infection was much higher (12.5%) in younger sex workers with age ≤ 20 years compared with older age groups (5.4%). In our series the age range was 22–66 years with a mean age of 38.4 years. Men were five times more commonly affected than women. The most common age group affected was the 31–40 years; this is consistent with the findings of Leynaert et al. [17] and Kumar [18] et al. who observed the highest incidence of HIV in the late twenties and thirties.

CD4+ T Cell Count

In our study, the mean CD4+ T cell count in the HIV group was 179 cells/ μ l. Rogers MC et al. [19] found the median CD4 count was 97 cells/ μ l amongst concordant men and 222 cells/ μ l amongst discordant men.

Organisms Isolated

Schmidt-Westhausen et al. [6] found that 5% of the HIV-infected patients and 4.8% of the controls had different species of *Enterobacteriaceae* in the oral cavity. Yeasts were always associated with *Enterobacteriaceae* in the HIV-infected patients. In our series the most common micro organism isolated in the control group was the Viridans streptococci (60%); this was followed by the *S. pneumoniae* in 23.33% cases. In the HIV group, we found a shift in oral microflora with a reduction in the isolation of Viridans streptococci (42%) and *S. pneumoniae* (6%). We found an increase in the incidence of colonization with *Micrococcus* spp. (22%). *Micrococcus* species are usually considered normal inhabitants of the skin [20] and the isolation and increased colonization of oral cavity with

these microorganisms has never been reported in the literature previously. *Micrococcus* species has been reported to cause localized cutaneous infections like folliculitis [20] and keratitis [21] in HIV-infected patients.

Gray et al. [5] stated that the HIV-infected individuals are predisposed to infection with bacteria like *S. pneumoniae*, non-typhi *Salmonella*, *S. aureus*, *Enterobacteriaceae*, *Rhodococcus* spp, *Nocardia* spp. Mascellino et al. [22] found high incidence of nosocomial bacterial infections in patients with AIDS and it was associated with high fatality rates. The most common micro-organisms isolated were gram-negative bacteria like *E. coli*, *Proteus*, *Enterobacter*, *Serratia*, and *Klebsiella*. The organisms that were isolated in our series were Viridans streptococci, *Micrococcus*, *Acinetobacter*, *Klebsiella* spp, *Pneumococcus*, *Bacillus*, and *Moraxella catarrhalis*. All these organisms are potentially pathogenic organisms especially so in a patient with HIV.

Viridans streptococci have been reported to cause various infections in HIV patients. Corti et al. [20] evaluated 61 episodes of infective endocarditis in intravenous drug abusers and human immunodeficiency type-1 virus infection. He isolated *S. aureus* in 73.1%, *S. viridans* in 19.5%, *S. epidermidis* in 2.4%, *S. hominis* in 2.4% and *S. pneumoniae* 2.4%. Milgrim et al. [21] studied sinusitis in human immunodeficiency virus infection and the most common organisms isolated were *S. pneumoniae* (19%), *S. viridans* (19%), and *Pseudomonas aeruginosa* (17%). It is important to recognize that infection due to atypical organisms must be considered if an HIV-infected patient with sinusitis does not respond to initial antibiotic therapy [22].

Acinetobacter has been reported to cause sepsis, urinary, respiratory tract diseases and bacteremic pneumonia [23, 24] in HIV-infected patients. As opportunist pathogens with predominant nosocomial origin *Acinetobacter* spp. organisms may be responsible for an appreciable morbidity in patients with HIV disease, especially when additional risk factors like immunodeficiency, underlying diseases, and hospitalization are present [23].

Correlation Between CD4+ T Cell Count and Microbiota in the HIV Group

Manfredi et al. [24] in their study on *Acinetobacter* infection in HIV patients found that all patients were severely immunocompromised with a mean CD4+ lymphocyte count of 122 cells/ μ l and a frequent prior diagnosis of AIDS. As opposed to other infections, septicaemia was associated with a significantly lower CD4+ T cell count and a more frequent occurrence of neutropenia. In our series, in patients with CD4+ T cell count less than 50 cells/ μ l we found a heavy colonization of the oral cavity with *Micrococcus* spp., *Acinetobacter* and *Klebsiella* spp.

As CD4+ T cells counts improved to 51–100 cells/ μ l, Viridans streptococcus colonies returned and 37.5% patients showed a heavy growth. *Micrococcus* spp. colonies were isolated till the CD4+ T cells improved up to 300 cells/ μ l. At counts >300 cells/ μ l the oral microbiota became comparable to that of the controls. A thorough search of available literature has failed to highlight the reason for such a shift in oral microbial flora with a change in CD4+ T cell count.

Antibiotic Sensitivity Testing

The antibiotic sensitivity patterns in both the groups were comparable. Although increased bacterial resistance to first line antibiotics has been reported by many authors [24, 25] we did not find any increase in the incidence of resistance. Surveillance studies of antimicrobial resistance have shown that bacterial resistance has been increasing in many nosocomial pathogens. Vancomycin-resistant *Enterococcus* increased in prevalence to 28.5%, whereas Methicillin Resistant *S. Aureus* had increased to 59.5%. Resistance of *K. pneumoniae* and *Enterobacter* spp. to third-generation cephalosporins increased to 20.6 and 31.1%, respectively. Resistance of *Pseudomonas aeruginosa* to imipenem, quinolones, and third-generation cephalosporin increased to 21.1, 29.5, and 31.9%, respectively [26]. Factors that increase antibiotic resistance include total antibiotic consumption as well as underuse through lack of access, inadequate dosing, poor adherence, and substandard antimicrobial usage [27].

Conclusion

Opportunistic infections are the most common cause of death in patients with HIV and many of these are caused by commensal bacteria which are otherwise harmless in a normal individual. Our study is unique in that such a study of the oral commensals in HIV patients has never been reported. We found an increased colonization of the oral cavity by *Micrococcus* spp. which is a normal commensal of the skin. We could also establish a shift in the normal flora with a decrease in the immune competence assessed by CD4+ T cell count. Although the exact significance of this phenomenon is not established, further research is required to ascertain its potential significance. There has been an alarming increase in the antibiotic resistance in HIV patients over the past few years. Although we did not find an increase in the resistance pattern, in wake of the numerous reports on antibiotic resistance we advice caution against the indiscriminate use of antibiotics.

Acknowledgment The authors thank the Indian Council of Medical Research for all the guidance and assistance in completing the research.

References

1. UNAIDS/WHO (2005) Joint United Nations Programme on HIV/AIDS (UNAIDS)
2. World Health Organization (WHO) (2005) AIDS epidemic update
3. HIV estimates (2004) NACO 2005
4. Overholser CD, Peterson DE, Williams LT, Schimpff SC (1982) Periodontal infection in patients with acute nonlymphocytic leukaemia: prevalence of acute exacerbations. *Arch Intern Med* 142:551–554
5. Gray KJ, French N (2004) Other bacteria and HIV disease. *Trop Dr* 34(4):206–208
6. Schmidt-Westhausen A, Fehrenbach FJ, Reichart PA (1990) Oral enterobacteriaceae in patients with HIV infection. *J Oral Pathol Med* 19(5):229–231
7. Nord CE, Kager L, Heimdahl A (1984) Impact of anti microbial agents on the gastrointestinal microflora and the risk of infection. *Am J Med* 76:99–106
8. Heimdahl A, Nord CE (1982) Effect of erythromycin and clindamycin on the indigenous human anaerobic flora and new colonization of the gastrointestinal tract. *Eur J Clin Microbiol* 1:38–48
9. Wahlin YB, Holm AK (1988) Changes in the oral microbiota in patients with acute leukemia and related disorders during the period of induction therapy. *Oral Surg Oral Med Oral Pathol* 65:411–417
10. Fauci AS, Lane HC (2001) Human immunodeficiency virus disease: AIDS and related disorders. In: Braunwald E, Hauser SL, Fauci AS, Longo DL, Kasper DL, Jameson JL (eds) *Harrison's principles of internal medicine*. McGraw-Hill, New York, pp 1876–1877
11. Mims C (2004) Diagnosis of infections and assessment of host defence mechanism. In: Mims C, Dockerell HM, Goering RV, Ivan R, Wakelin D, Zuckerman M (eds) *Medical microbiology*. Elsevier, London, pp 453–471
12. Chaisson RE (1988) Infections due to encapsulated bacteria, *Salmonella*, *Shigella*, and *Campylobacter*. *Infect Dis Clin North Am* 2:475–484
13. Krasinski K (1994) Bacterial infections. In: Pizzo PA (ed) *Pediatric AIDS*. Williams & Wilkins, Baltimore, pp 241–253
14. Wilfert CM (2000) Invasive bacterial infections in children with HIV infection. In: Pizzo PA, Wilfert CM (eds) *Pediatric AIDS*. Williams & Wilkins, Baltimore, pp 117–124
15. Cruciani M, Luzzati R, Fioredda F, Loy A, Molesini M, Concia E, Jannuzzi C, Bassetti D (1993) Mucosal colonization by pyogenic bacteria among children with HIV infection. *Aids* 7:1533–1534
16. Sarkar K, Bal B, Mukherjee R, Saha MK, Chakraborty S, Niyogi SK, Bhattacharya SK (2005) Young age is a risk factor for HIV among female sex workers: an experience from India. *J Infect* (Epub ahead of print)
17. Leynaert B, Downs AM, de Vincenzi I (1998) Heterosexual transmission of human immunodeficiency virus: variability of infectivity throughout the course of infection. European study group on heterosexual transmission of HIV. *Am J Epidemiol* 148(1):88–96
18. Kumar R, Jha P, Arora P, Mony P, Bhatia P, Millson P, Dhingra N, Bhattacharya M, Remis RS, Nagelkerke N (2006) International studies of HIV/AIDS (ISHA) investigators. Trends in HIV-1 in young adults in south India from 2000 to 2004: a prevalence study. *Lancet* 367(9517):1164–1172
19. Rogers MC, Gopalakrishnan G, Kumarasamy N, Flanigan TP, Carpenter CC, Mayer KH, Solomon S (2005) HIV in couples in South India; implications for prevention. *Int J STD AIDS* 16(6):442–445

20. Corti ME, Palmieri OJ, Villafane MF, Trione N (2004) Evaluation of 61 episodes of infective endocarditis in intravenous drug abusers and human immunodeficiency type-1 virus infection. *Rev Argent Microbiol* 36(2):85–87
21. Milgrim LM, Rubin JS, Rosenstreich DL, Small CB (1994) Sinusitis in human immunodeficiency virus infection: typical and atypical organisms. *J Otolaryngol* 23(6):450–453
22. Mascellino MT, De Sanctis GM, Iegri F, De Gregoris P, Sorice F (1989) Significant bacteriological findings in HIV-positive patients. *Boll Ist Sieroter Milan* 68(3):271–276
23. Manfredi R, Nanetti A, Valentini R, Chiodo F (2001) Pathogenic role of *Acinetobacter* spp. during HIV infection. *Infez Med* 9(1):43–51
24. Manfredi R, Nanetti A, Valentini R, Chiodo F (2001) *Acinetobacter* infections in patients with human immunodeficiency virus infection: microbiological and clinical epidemiology. *Chemotherapy* 47(1):19–28
25. Molyneux E (2004) Bacterial infections in children with HIV/AIDS. *Trop Dr* 34(4):195–198
26. National nosocomial infections surveillance (NNIS) (2004) System report, data summary from January 1992 through June 2004, issued October 2004. *Am J Infect Control* 32:470–85
27. Spector SA, Gelber RD, McGrath N (1994) A controlled trial of intravenous immune globulin for the prevention of serious bacterial infections in children receiving zidovudine for advanced HIV infection. *N Engl J Med* 331:1181–1187