Clinical and Microbiological Effects of Levofloxacin in the Treatment of *Aggregatibacter actinomycetemcomitans*-associated Periodontitis: A Randomized Placebo-Controlled Clinical Trial

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Abstract

Aim: To evaluate the clinical and microbiological effects of systemic levofloxacin (LFX) in subjects with *Aggregatibacter actinomycetemcomitans*-associated chronic periodontitis (AA-ACP).

Materials and methods: Subjects with severe periodontitis with subgingival detection of *A. actinomycetemcomitans* were randomly divided into two treatment groups; a test group (n = 35) that received scaling and root planing (SRP) and LFX (500 mg o.d.) and a control group (n = 34) that received SRP and placebo (o.d.) for 10 days. Plaque index (PI), gingival index (GI), percent of sites with bleeding on probing (%BoP), probing depth (PD) and clinical attachment level (CAL) were recorded and subgingival plaque samples were cultivated for detection of *A. actinomycetemcomitans* at baseline to 6 months at various intervals.

Results: Subjects receiving LFX showed the greatest improvements in mean PD and CAL. The difference in the reduction of PD and CAL in the two groups was significant at 1, 3 and 6 months for PD and 3 and 6 months for CAL ($p < 0.05$). The inter-group difference in PI, GI and %BoP was not significant at any interval. Detectable levels of *A. actinomycetemcomitans* were significantly less in the test group 3 and 6 months post-therapy.

Conclusion: Systemic LFX as an adjunct to SRP improves clinical outcomes and suppresses *A. actinomycetemcomitans* below detectable levels.

Key words: Antibiotics, levofloxacin, periodontitis, subgingival plaque

Introduction

Periodontitis is chronic infection that results from disturbance in equilibrium between the host defense mechanism and periodontopathic microorganisms and causes progressive destruction of the periodontal tissues. The progression of periodontitis is closely related to the colonization of microorganisms, including *Aggregatibacter actinomycetemcomitans* (previously *Actinobacillus actinomycetemcomitans*), *Porphyromonas gingivalis*, *Tannerella forsythia*, and *Treponema denticola* (Genco *et al.,* 1996).

A. actinomycetemcomitans was first isolated by Klinger in actinomycotic mixed infections (Klinger, 1912). Later, *Actinobacillus actinomycetemcomitans*, *Haemophilus aphrophilus*, *Haemophilus paraphrophilus* and *Haemophilus segnis* were reclassified as *Aggregatibacter actinomycetemcomitans* (Norskov-Lauritsen and Kilian, 2006). It is a microaerophilic, non-motile, Gram-negative capnophilic coccobacillus that plays a major role in the etiology of localized aggressive periodontitis and is also thought to be associated with the etiology of generalized aggressive periodontitis and chronic periodontitis (Klinger, 1912; Tanner *et al.,* 1979; Zambon *et al.,* 1983; Slots, 1986; Preus *et al.,* 1987; Christersson *et al.,* 1992). Prevention and control of *A.*

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actinomycetemcomitans-associated chronic periodontitis (AA-ACP) is challenging. Although nonsurgical periodontal therapy (NSPT) is effective treatment of chronic periodontitis, it seems less effective in patients with AA-ACP and often is not able to suppress *A. actinomycetemcomitans* below detectable level (Slots and Rosling, 1983; Renvert *et al.,* 1990; Renvert *et al.,* 1990; AAP, 2000). Antibiotics are a useful aid in the treatment of periodontitis. The effectiveness of systemic antibiotics as adjuncts to scaling and root planing (SRP) has been investigated, including tetracycline (e.g., doxycycline), amoxicillin, metronidazole, and various combinations of these. Most of the studies show improved clinical outcomes with the adjunctive use of antibiotics (Herrera *et al.,* 2002; Slots and Ting, 2002; Walker and Karpinia, 2002; Haffajee *et al.,* 2003; Ribeiro Edel *et al.,* 2009; Rodrigues *et al.,* 2012), but some of the studies have shown contradictory results (Saxen *et al.,* 1990; Saxen and Asikainen, 1993; Palmer *et al.,* 1996; Tinoco *et al.,* 1998).

Antibiotics that have been studied most in treatment of periodontal diseases include tetracyclines and metronidazole, frequently combined with amoxicillin (Walker and Karpinia, 2002). Drawbacks associated with these drug regimens are a decrease in compliance because of the need to take many pills a day (Greenberg, 1984), and increase in bacterial resistance (Villedieu *et al.,* 2003; Al-Haroni *et al.,* 2006). Results of some studies have shown that systemic tetracyclines along with NSPT is able to improve treatment outcome in some cases, but this treatment mostly fails to eliminate *A. actinomycetemcomitans* from subgingival areas (Slots and Rosling, 1983; Mandell *et al.,* 1986; Mandell and Socransky, 1988; Christersson and Zambon, 1993). A recent study (Mombelli *et al.,* 2013) has shown that *A. actinomycetemcomitans*-positive patients had no specific benefit from amoxicillin and metronidazole.

Because of the above-mentioned drawbacks of established antibiotics there is a need to investigate the effects of other antibiotics without these disadvantages against *A. actinomycetemcomitans*. Fluoroquinolones could be a better alternative as these are effective against the *Pasteurellaceae* family to which *A. actinomycetemcomitans* belongs (Tanner *et al.,* 1994). Some studies have shown good results with adjunctive use of fluoroquinolones such as ciprofloxacin and ofloxacin in AA-ACP (Kleinfelder *et al.,* 2000; Naokotakahashi *et al.,* 2007; Suci and Young, 2011). Ciprofloxacin significantly reduced the microcolony size and cell surface density of *A. actinomycetemcomitans* in the dual species biofilm over a 24-hour period (Suci and Young, 2011). Ofloxacin exerted a strong inhibitory effect in both the early and mature phases of *A. actinomycetemcomitans* biofilm formation (Naokotakahashi *et al.,* 2007).

Levofloxacin (LFX) is the synthetic *L*-isomer of the racemic quinolone ofloxacin and is active against a broad range of Gram-positive, Gram-negative and atypical bacteria (Anderson and Perry, 2008). LFX showed advantages over ciprofloxacin in terms of clinical efficacy and disease recurrence, with a low rate of adverse events, for the treatment of chronic bacterial prostatitis (Zhang *et al.,* 2012). LFX was found successful in all patients suffering from invasive *A. actinomycetemcomitans* infection, and none of the patients demonstrated recurrence (Wang *et al.,* 2010).

To date, no study has ever tested LFX in the treatment of periodontitis. The aim of the present study was to evaluate the clinical and antimicrobial effect of systemic LFX as an adjunct to SRP in the treatment of subjects with AA-ACP.

Materials and methods

Subjects

Sixty-nine subjects with a history of severe periodontitis (Armitage, 1999) harbouring *A. actinomycetemcomitans* were selected from the population referred to the Department of Periodontics, Government Dental College and Research Institute (GDCRI), Bangalore between February and September of 2012. The research protocol was initially submitted and approved by the Institutional Ethical Committee and Review Board of the Government Dental College and Research Institute, Bangalore. After ethical approval, all subjects were verbally informed and written informed consent was taken for participation in the study.
[IRB no: GDCRI/ACM/PG/PhD/2/2011-2012]

Inclusion and exclusion criteria

After a detailed medical history and an initial clinical and radiological examination (intra-oral periapical radiograph using paralleling technique), systemically healthy subjects with previously untreated severe periodontitis were selected.

Inclusion criteria were a minimum of 20 teeth, subgingival detection of *A. actinomycetemcomitans*, periodontal bone loss around at least two teeth per quadrant reaching the middle third of the roots, with probing depths (PD) of at least 6 mm and clinical attachment level (CAL) of at least 7 mm. Patients with use of systemic antibiotics in the previous 6 months, known systemic disease, known allergy to fluoroquinolones, alcoholics, immunocompromised subjects and pregnant or lactating females were excluded from the study.

Sample size calculation

The ideal sample size to ensure adequate power for this clinical trial was calculated considering differences of at least 1 mm between groups for clinical attachment level (CAL), changes in sites with initial probing depth (PD) $\geq$ 6mm and assuming a standard deviation of 1.0 mm (Matarazzo *et al.,* 2008). Based on these calculations, it was defined that 18 subjects per group would be necessary to provide an 80% power with an α of 0.05.

Study design and treatment protocol

A double-blinded, randomized, placebo-controlled clinical trial was designed. A total of 87 subjects were assessed for eligibility. Of these 87, 69 subjects meeting the inclusion criteria were randomly assigned to one of the two groups using a computer-generated random allocation sequence by the chief coordinator (ARP). Three of the 69 selected subjects (one from Group 1 and two from Group 2) did not return for the 3-month follow-up visit and were excluded from the statistical analysis.

Group 1 (Test: SRP+ LFX 500 mg (Blee 500 mg tablets, Schwitz Biotech Pvt Ltd., Ahmedabad, India) o.d. x 10 days) had 34 subjects (16 female and 18 male, mean age 36.7 ± 6.2 years) while Group 2 (Control: SRP + Placebo x 10 days) comprised 32 subjects (16 female and 16 male, mean age 36.8 ± 5.8). A placebo that looked similar to LFX was prepared at Government College of Pharmacy, Bangalore. These medications were put in brown coloured opaque packets marked with only the subject number by the study coordinator.

Supragingival scaling was performed on all the participants one week before the baseline visit. Strict oral hygiene instructions were given to all participants at the same time by an experienced dentist (SSM). Participants were also instructed to use 0.2% chlorhexidine mouthrinse twice daily during this period. A clinical examiner calibration exercise was performed on 15 patients, 48 hours apart, and the difference between the measurements was within 1 mm in 95% of measurements for PD and CAL.

At the baseline visit, the clinical parameters were recorded and plaque samples were collected from each patient in both groups by the clinical examiner (SPS). Next, the dentist (SSM) performed thorough SRP under local anesthesia for all the participants. After this, each participant received a package containing the test or placebo medication by the clinical examiner, who was blinded to the contents of the packets. All packages were identical in appearance and were marked only with the participant number. All subjects were carefully informed about medication intake. The subjects were asked to bring the packets at the end of the 10-day regimen to check for compliance.

The treatment group was masked from the patient, clinical examiner, operator and statistician throughout the duration of the study.

Clinical recordings

The clinical parameters to be recorded included: plaque index (PI; Quigley and Hein, 1962; Turskey *et al.,* 1970), gingival index (GI; Loe and Sillness, 1963), percent of sites with bleeding on probing (%BoP), PD and CAL.

The CAL was measured to the nearest mm from the cementoenamel junction (CEJ) to the deepest probeable point (Glavind and Loe, 1967) using a standardized periodontal probe (UNC 15 periodontal probe, Hu-Friedy, IL, USA), and PD was measured from the gingival margin to the bottom of the pocket. Bleeding on probing was scored positive if a site bled immediately after pocket probing or if a site bled at completion of the probing of a jaw quadrant.

These parameters were recorded in all teeth excluding third molars (6 sites/tooth). These 6 sites included mesio-buccal, mid-buccal, disto-buccal, mesio-palatal/ lingual, midpalatal/ lingual and disto palatal/lingual. PI and GI were recorded before instituting supragingival scaling at baseline.

Clinical measurements were recorded at baseline, 10 days, 1 month, 3 months and 6 months following therapy.

Microbiological analysis

Microbiological analysis was performed in all patients at baseline, 3 months and 6 months after therapy. Intracrevicular *A. actinomycetemcomitans* was evaluated in subgingival plaque samples. These samples were collected from the same sites throughout the study at the deepest site of each quadrant showing PD ≥ 6 mm and CAL ≥ 7 mm (Savitt *et al.,* 1991). Sterile endodontic absorbent points were inserted at the bottom of the periodontal pocket for 10 seconds to collect subgingival plaque samples. Four samples from each patient were transferred to 1 ml anaerobically prepared and stored sterile half-strength Ringer's solution. The pooled samples were sonicated for 10 seconds and diluted in 10-fold steps, 0.1 ml of the undiluted suspension and 0.1 ml aliquots of dilutions were spread on freshly prepared tryptic soy-serum-bacitracin-vancomycin (TSBV) agar plates (Slots, 1982). Plates were incubated for 3 days at 36°C in an atmosphere containing 85% N_2, 10% H_2 and 5% CO_2. *A. actinomycetemcomitans* was identified on TSBV agar as small convex colonies with a star-like inner structure and positive catalase reaction. Optimum dilution plates with 50 to 150 colonies were selected and the total number of colony forming units (CFU) was enumerated. The number of CFU was calculated for 1 ml undiluted Ringer's solution (Kleinfelder *et al.,* 2000).

Primary and secondary outcome measures

The primary outcome variable was the differences in the test and control groups for the mean CAL change from baseline to 10 days, 1, 3 and 6 months. The secondary outcome measures were the differences in both the groups for mean reduction in PD, PI, GI, %BOP and reduction in detectable levels of *A. actinomycetemcomitans* from baseline to subsequent intervals.

Statistical analysis

Statistical analysis was performed by statistical software (SPSS, version 14.0). Independent samples *t*-tests were used to compare PD, CAL, GI and PI between test and

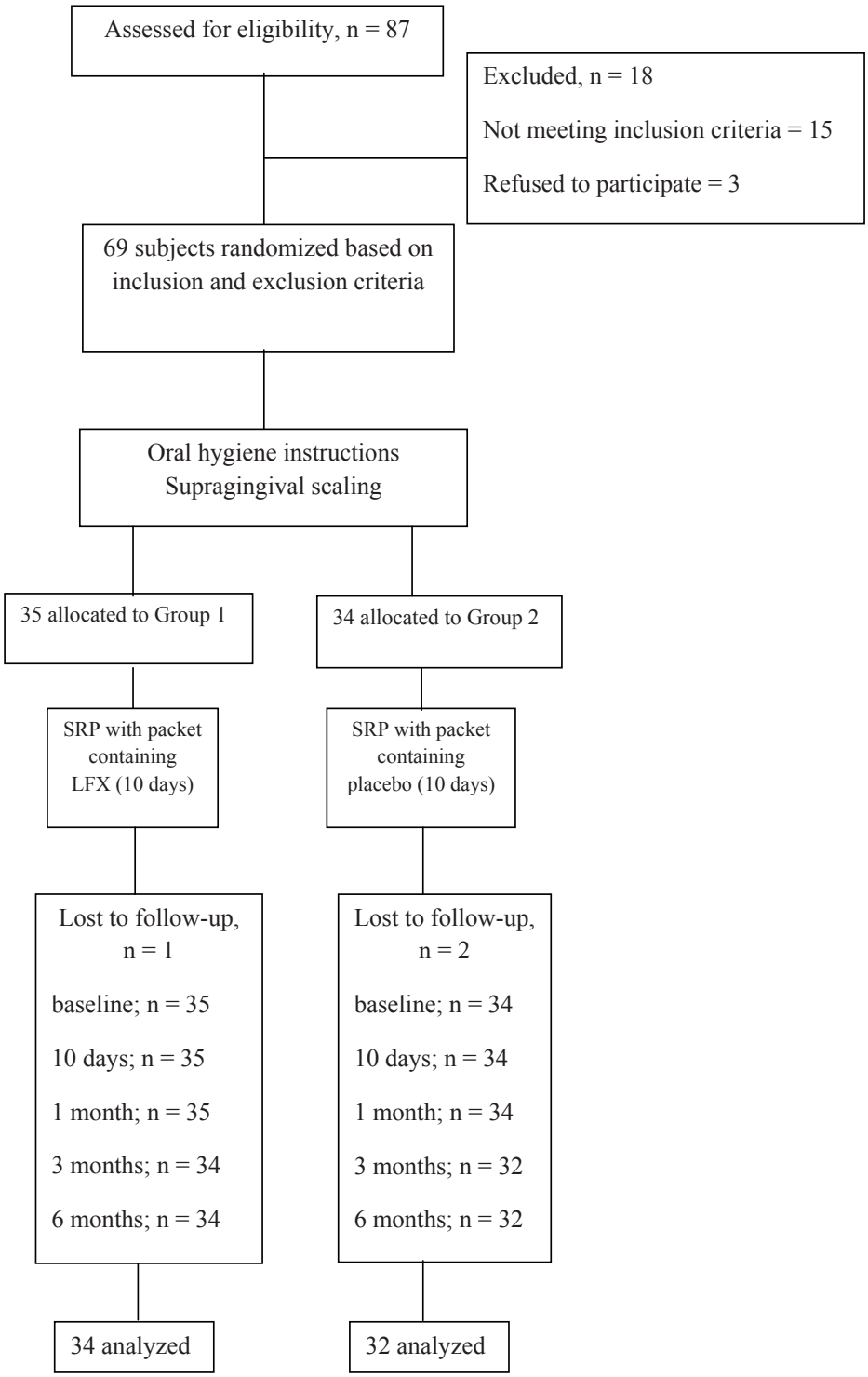

Figure 1. CONSORT flow chart

control groups. To compare before and after treatment data of these parameters within test and control groups, Student's *t*-test for paired samples was used. Repeated measures ANOVA was used for comparison of PD and CAL between the two groups and to assess the change in PD and CAL at all time intervals for the within- and between-subject effect. To compare BoP data between both groups the Mann-Whitney U test was employed, and to compare before and after treatment data within groups, Wilcoxon's rank test was employed. Fisher's exact test was used to compare microbiological findings between test and control groups.

Results

Clinical measurements

Sixty-six subjects could be evaluated finally at the end of 6 months. *Figure 1* shows the CONSORT diagram describing the number of subjects analyzed and those dropping out of the study. *Table 1* gives the mean PD

and CAL values in the two groups at baseline, 10 days, 1 month, 3 months and 6 months. *Table 2* describes the change in PD and CAL at all time intervals with the within-subject effect by repeated measures ANOVA. *Figure 2* and *Figure* 3 are line graphs of PD and CAL values, respectively, at various intervals. There was a reduction in PD at 10 days, but the difference between the two groups was not statistically significant. The difference in mean PD values between the two groups at months 1, 3 and 6 was statistically significant ($p < 0.05$). The mean CAL value also showed a statistically significant difference at 3 months ($p < 0.0001$; 95% CI: -1.65 to -0.80) and 6 months ($p = < 0.0001$; 95% CI: -1.74 to -0.92).

The difference in GI, PI and %BoP was not statistically significant at any time point ($p > 0.05$) as shown in *Table 3* and *Table 4*. The intra-group differences in all the clinical parameters from baseline to various time intervals were statistically significant.

Microbiological analysis

In *Table 5* the number of CFUs of *A. actinomycetemcomitans* at baseline, 3 months and 6 months is shown in logarithmic counts (first quartile, median and third quartile values) for test and control group subjects. *A. actinomycetemcomitans* was present at baseline in all subgingival samples of test and control subjects. As

Table 1. Mean value (standard deviation), *p*-values and confidence interval (CI) of probing (PD) depth and clinical attachment level (CAL).

		Test	Control	*p* - value	CI
PD (mm)	Baseline	6.66 (1.18)	6.54 (1.18)	0.665	-0.45 to 0.69
	10 days	5.64 (0.90)	5.53 (0.88)	0.599	-0.32 to 0.56
	1 month	4.79 (0.70)	5.17 (0.80)	0.0413*	-0.75 to -0.02
	3 months	4.05 (0.60)	5.25 (0.83)	< 0.0001*	-1.55 to -0.84
	6 months	4.04 (0.54)	5.33 (0.83)	< 0.0001*	-1.63 to -0.95
CAL (mm)	Baseline	7.82 (1.32)	7.58 (1.20)	0.4324	-0.38 to 0.87
	10 days	6.75 (1.13)	6.56 (0.95)	0.4762	-0.33 to 0.70
	1 month	5.81 (0.95)	6.17 (0.87)	0.1091	-0.82 to 0.08
	3 months	5.05 (0.81)	6.27 (0.91)	< 0.0001*	-1.65 to -0.80
	6 months	5.05 (0.75)	6.38 (0.92)	< 0.0001*	-1.74 to -0.92

*Statistically significant at 5% level of significance ($p < 0.05$).

Table 2. Change in probing depth (PD) and clinical attachment level (CAL) at all time intervals with the within-subject effect by repeated measures analysis of variance (ANOVA)

Source	Type III sum of squares	df	Mean square	F value	*p* - value
			(PD)		
Test of within-subjects effect					
Time	175.74	4	43.94	507.82	< 0.0001*
Time x group	31.10	4	7.77	89.86	< 0.0001*
Error (time)	22.15	256	0.087		
Test of between-subjects effects					
Group	22.86	1	22.86	6.84	0.01*
Error	214.05	64	3.345		
			(CAL)		
Test of within-subjects effect					
Time	192.47	4	48.12	569.52	< 0.0001*
Time x group	37.24	4	9.31	110.18	< 0.0001*
Error (time)	21.63	256	0.84		
Test of between-subjects effects					
Group	20.47	1	20.47	4.44	0.04*
Error	295.35	64	4.62		

*Statistically significant at 5% level of significance ($p < 0.05$).

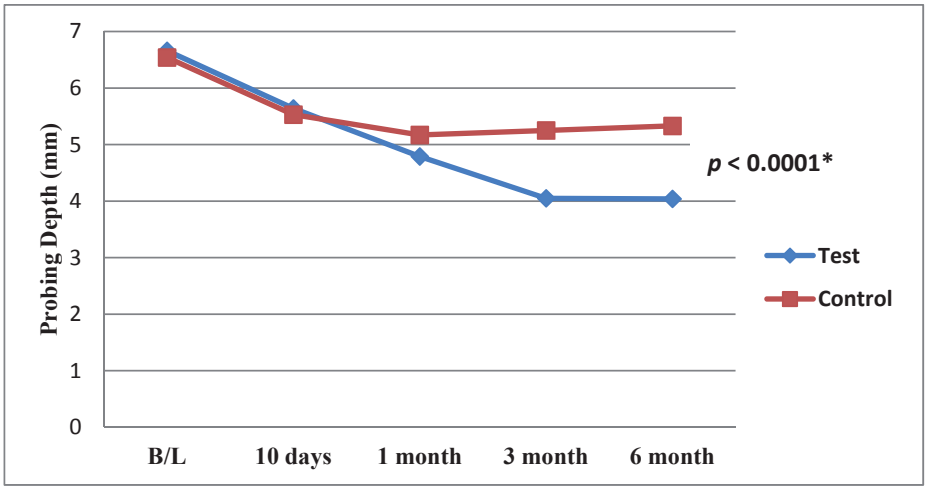

Figure 2. Probing depth values (mm) at various intervals. B/L, baseline.

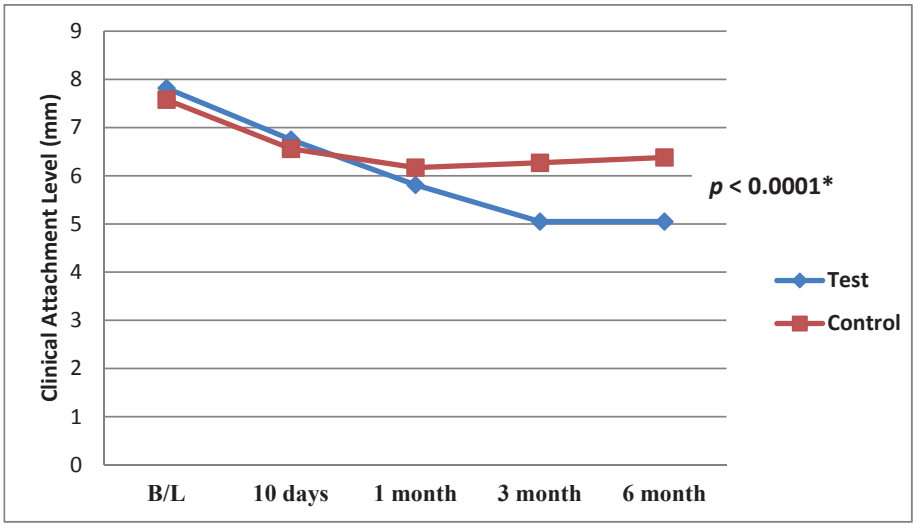

Figure 3. Clinical attachment level (mm) values at various intervals. B/L, baseline.

shown in *Table 6*, *A. actinomycetemcomitans* was found to be below detectable levels in 32/34 subjects at the 3 months examination and in 31/34 subjects at the 6 months examination in the test group; whereas in control group, *A. actinomycetemcomitans* was recovered in 24 and 28 of 32 control subjects at the 3-month and 6-month examinations, respectively.

Adverse drug reactions

Two participants (one male and one female) in the test group reported dizziness. Another female patient in the same group complained of diarrhea.

Discussion

The aim of the present randomized, controlled trial was to assess the effect of adjunctive use of LFX with non-surgical treatment of AA-ACP. Scaling and root planing combined with a 10-day regimen of LFX resulted in significantly better clinical outcome parameters, including PD and CAL, compared with SRP combined with placebo. In addition, significant differences in microbiological outcomes were also detected: at the 6-month examination subgingival *A. actinomycetemcomitans* was found to be below detectable levels in 91% of the subjects in the test group as compared to only 12% of subjects in the control group. To the best of our knowledge, this is the first study showing the clinical efficacy of LFX 6 months following non-surgical treatment of AA-ACP.

There are numerous studies on systemic use of antibiotics as an adjunct to SRP. Some of the earlier studies with the use of antibiotics as an adjunct to NSPT were not able to demonstrate a benefit in results (Lindhe *et al.,* 1983; Saxen *et al.,* 1990; Saxen and Asikainen, 1993; Palmer *et al.,* 1996; Tinoco *et al.,* 1998). Other studies supported the clinical benefits of using adjunctive antibiotics in the treatment of periodontal disease (Herrera *et al.,* 2002; Slots and Ting, 2002; Walker and Karpinia,

Table 3. Mean value (standard deviation), *p* - values and confidence intervals (CI) of gingival index (GI) and plaque index (PI).

		Test	Control	*p* - value	CI
GI	Baseline	2.15 (0.27)	2.13 (0.31)	0.7941	-0.12 to 0.16
	10 days	1.46 (0.28)	1.38 (0.29)	0.2157	-0.05 to 0.23
	1 month	1.34 (0.26)	1.33 (0.23)	0.8329	-0.11 to 0.13
	3 months	1.33 (0.25)	1.24 (0.15)	0.110	-0.02 to 0.19
	6 months	1.40 (0.28)	1.29 (0.18)	0.0665	-0.01 to 0.23
PI	Baseline	3.58 (0.54)	0.54 (0.55)	0.8991	-0.25 to 0.29
	10 days	2.43 (0.44)	2.47 (0.41)	0.7299	-0.25 to 0.17
	1 month	2.37 (0.48)	2.36 (0.37)	0.9611	-0.21 to 0.22
	3 months	2.49 (0.49)	2.49 (0.37)	0.9973	-0.22 to 0.22
	6 months	2.62 (0.50)	2.65 (0.37)	0.7894	-0.25 to 0.19

Table 4. Mean value (standard deviation), *p*-values and U values of bleeding on probing (BoP) sites.

		Test	Control	*p* - value	U
%BoP	Baseline	65.8 (11.33)	66.6 (12.87)	> 0.05	587
	10 days	28.4 (6.01)	29.1 (9.27)	> 0.05	573.5
	1 month	16.8 (5.37)	17.9 (6.32)	> 0.05	633
	3 months	19.1 (5.86)	19.9 (6.27)	> 0.05	603
	6 months	22.0 (6.29)	22.8 (6.45)	> 0.05	588.5

Table 5. Colony forming units (CFU) of *Aggregatibacter actinomycetemcomitans* (Aa)in logarithmic counts (first quartile, median and third quartile values)

	Test Log *Aa*			Control Log *Aa*		
Quartile	**baseline**	**3 months**	**6 months**	**baseline**	**3 months**	**6 months**
I quartile (Lower)	4	0	0	4	0.5	2.25
Median	5	0	0	5	2	4
III quartile (Upper)	5	0	0	5	3	4

Table 6. Number and percent of patients with subgingival detection of *Aggregatibacter actinomycetemcomitans*

	Test	Control
Baseline	34/34 (100)	32/32 (100)
3 months	2/34 (5.9)*	24/32 (75)
6 months	3/34 (8.8)*	28/32 (87.5)

*Statistically significant difference between test and control group ($p < 0.0001$)

2002; Haffajee *et al.,* 2003; Ribeiro Edel *et al.,* 2009; Rodrigues *et al.,* 2012). Studies have shown beneficial results with systemic tetracyclines or amoxicillin - metronidazole along with NSPT in some cases, but this treatment mostly fails to eliminate *A. actinomycetemcomitans* from subgingival areas (Slots and Rosling, 1983; Mandell *et al.,* 1986; Mandell and Socransky, 1988; Christersson and Zambon, 1993) or to show any additional benefit in patients harbouring *A. actinomycetemcomitans* (Mombelli *et al.,* 2013).

Doxycycline has shown beneficial results in *A. actinomycetemcomitan*-associated localized aggressive periodontitis in six subjects (Mandell *et al.,* 1988). Doxycycline and metronidazole were also found effective in treatment of subjects with recurrent chronic periodontitis (Lundstrom *et al.,* 1984; McCulloch *et al.,* 1990; Aitken *et al.,* 1992). Another clinical study investigated the effects of different antimicrobials on clinical outcome in subjects with generalized aggressive periodontitis, and in this study, doxycycline did not demonstrate any additional benefit in the reduction of deep pockets (>6 mm) as compared to NSPT alone (Xajigeorgiou *et al.,* 2006).

As most of the antibiotics that have been studied in the treatment of periodontal diseases demonstrated conflicting results or an increase in bacterial resistance (Al-Haroni *et al.,* 2006; Villedieu *et al.,* 2003), there is a need to investigate other antibiotics effective against *A. actinomycetemcomitans*.

Quinolones were introduced for use in urinary tract infections in the 1960s (Reese, 1965). They represent one of the main classes of antibiotics. Advantages of quinolones are good penetration into tissues and antibacterial activity within cells (Van Bambeke *et al.,* 2005).

Older generation fluoroquinolones such as ciprofloxacin and ofloxacin have shown good results when used as an adjunct in AA-ACP (Kleinfelder *et al.,* 2000; Naokotakahashi *et al.,* 2007; Suci and Young, 2011). *A. actinomycetemcomitans* residing in a dual species biofilm with the commensal *S. sanguis* can be selectively killed, or at least rendered metabolically inactive, by treatment with ciprofloxacin (Suci and Young, 2011). In a study on 25 adult periodontitis patients it has been shown that systemic ofloxacin as an adjunct to flap surgery is able to suppress *A. actinomycetemcomitans* (Kleinfelder *et al.,* 2000). A strong inhibitory effect of ofloxacin has been demonstrated in both the early and mature phases of *A. actinomycetemcomitans* biofilm formation (Naokotakahashi *et al.,* 2007).

Levofloxacin, a newer generation fluoroquinolone, is the isomer of ofloxacin. Following oral administration, LFX is rapidly absorbed and maximum plasma concentrations are attained in 1–2 hours (Croom and Goa, 2003). Older generation quinolones act only on Gram-negative aerobic bacteria (Van Bambeke *et al.,* 2005), whereas LFX is active against a broad range of Gram-positive, Gram-negative and atypical bacteria (Anderson and Perry, 2008). Levofloxacin is distributed extensively in tissues and fluids throughout the body (Langtry and Lamb, 1998) and accumulates in phagocytic cells (Croom and Goa, 2003). At a lower dosage of LFX (500 mg once daily for 3 days), maximum plasma concentration (C_{max}) and area under the plasma concentration time curve from time 0 to 24 hrs (AUC24) values for the drug were significantly ($p < 0.01$) higher in the polymorphonuclear leukocytes than in plasma (Garraffo *et al.,* 2005). Levofloxacin is rapidly absorbed after oral administration and shows linear pharmacokinetics for both single- and multiple-dose (once daily) regimens. The oral solution and tablet formulations are bioequivalent to the intravenous formulation (Croom and Goa, 2003). After oral administration, the C_{max} of LFX is reached within 1–2 hours with an absolute bioavailability of oral LFX 500 mg of approximately 99% (Chien *et al.,* 1998; Chow *et al.*, 2001; Croom and Goa, 2003).

The oral dose of LFX is 500 mg once daily for 10 days, as used normally in community-acquired pneumonia, acute bacterial sinusitis and urinary tract infections (Anderson and Perry, 2008). Hence, less frequent dosing of LFX is required as compared to other antibiotics. Levofloxacin was found to be successful in patients suffering from invasive *A. actinomycetemcomitans* infection, and none of the patients demonstrated recurrence (Wang *et al.,* 2010). In an *in vitro* study LFX and ciprofloxacin showed high-potency antibacterial activity against clinically isolated *A. actinomycetemcomitans* (MIC90 0.013-0.025 μg/ml) (Equchi *et al.,* 2002). In a study on Chinese patients LFX showed advantages over ciprofloxacin in terms of clinical efficacy and disease recurrence, with a low rate of adverse events, for the treatment of chronic bacterial prostatitis (Zhang *et al.,* 2012).

Although LFX is a very safe fluoroquinolone, caution and a risk/benefit assessment is required when used in the elderly because of the increased risk of severe tendon disorders in this group of patients, particularly if they are receiving corticosteroids (Raritan, 2009). However, it should be stated that there is no evidence that tendon rupture is more likely to occur with LFX than with any other fluoroquinolone (*The Medical Letter on Drugs and Therapeutics,* 2011). Blood glucose monitoring is recommended in patients with diabetes mellitus receiving simultaneous hypoglycemic agents and/or insulin, because symptomatic hyperglycemia and hypoglycemia have been reported with LFX administration (Raritan NJ 2009). Concomitant administration of fluoroquinolones (including LFX) with non-steroidal anti-inflammatory drugs may increase the risk of central nervous system stimulation and convulsive seizures (Raritan, 2009). Levofloxacin should not be used in pediatric patients aged <18 years; the incidence of musculoskeletal disorders was shown to be higher in LFX-treated children (aged <5 years) than in those treated with a non-LFX therapy (Raritan, 2009).

To the best of our knowledge, this is the first study in which LFX has been evaluated as an adjunct to SRP in patients with a specific microbiological profile. Other authors have previously followed the same strategy with other antimicrobials (*i.e.*, selection of patients with a specific microbiological profile) in order to prescribe an appropriate drug: presence of *A. actinomycetemcomitans* or *P. gingivalis* when assessing amoxicillin plus metronidazole (Flemmig *et al.,* 1998); presence of *A. actinomycetemcomitans* when assessing ofloxacin (Kleinfelder *et al.,* 2000); or presence of *A. actinomycetemcomitans* and metronidazole in localized juvenile periodontitis (Saxen and Asikainen, 1993). All the above-mentioned studies and our study have in common excellent outcomes in the test groups, suggesting better results of adjunctive systemic antibiotics if the target pathogen has been identified previously (Herrera *et al.,* 2002).

Uncontrolled use of antimicrobials is of great health concern because of increasing bacterial resistance,

resulting in different bacterial antibiotic susceptibility profiles in different European countries according to greater or lesser prescription control (van Winkelhoff *et al.,* 2005). In order to optimize the use of antimicrobials to only those subjects who would benefit most, all the subjects included in the present study harbored *A. actinomycetemcomitans*. After treatment, *A. actinomycetemcomitans* detection was significantly reduced in the test group at 3 and 6 months; conversely, in the control group, the decrease in *A. actinomycetemcomitans* was less pronounced.

Although improved clinical and microbiological outcomes were attained in the test group using adjunctive LFX, limited microbiological analysis was the major limitation. These results make the use of this antibiotic recommendable in the treatment of periodontitis patients harboring *A. actinomycetemcomitans*.

Conclusion

We conclude that within the limitations of the present study, patients with advanced periodontitis harboring *A. actinomycetemcomitans* in their subgingival biofilm may benefit from the systemic administration of LFX as an adjunct to SRP. However, further long-term longitudinal trials considering other periodontopathic microorganisms are required to confirm the effectiveness of LFX in periodontitis.

Acknowledgments

We are thankful to Schwitz Biotech Pvt. Ltd., Ahmedabad, India for providing us levofloxacin (Blee) 500 mg tablets and the Government College of Pharmacy, Bangalore, for preparing placebos.

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ERRATUM

We have been advised that the affiliations of the authors were not correctly listed in the paper entitled "Tooth Loss Assessment during Periodontal Maintenance in Erratic versus Complete Compliance in a Periodontal Private Practice in Shiraz, Iran: A 10-Year Retrospective Study" published in the *Journal of the International Academy of Periodontology* 2014; **16**:43-49. The correct affiliations are as follows:

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