Effects of Metronidazole Plus Amoxicillin in Progressive Untreated Adult Periodontitis: Results of a Single 1-Week Course After 2 and 4 Months*

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A RANDOMIZED, DOUBLE-BLIND, CLINICAL STUDY was done to assess the microbiological and clinical effects of metronidazole plus amoxicillin (M+A) as the only therapy in 46 patients with moderate to advanced progressive adult periodontitis. Patients were included in the study after at least 2 sites showed ≥2 mm clinical attachment loss. Bleeding on probing, probing depth, and clinical attachment level were measured using an automated probe. The percentage of surfaces with plaque was recorded at day 0, and at 2 and 4 months after therapy. No effort was made to change the oral hygiene habits of patients. Identification of *Actinobacillus actinomycetemcomitans, Porphyromonas gingivalis,* and *Prevotella intermedia* was assessed utilizing DNA technology at day 0 and 2 months after therapy. Twenty-three patients received metronidazole 250 mg plus amoxicillin 500 mg, 3 times/day for a week and 23 a placebo. Two patients in the placebo group were dropped at 2 months because they had taken antibiotics for medical reasons. Statistical analyses of differences between groups was done using the Mann-Whitney test, and the differences within each group were tested with ANOVA. There were no significant changes in surfaces with plaque in either group after therapy. The percentage of bleeding sites decreased significantly from baseline to 2 and 4 months in the M+A group ($P = 0.001$), and increased in the placebo group. Differences in bleeding on probing between groups were significant at 2 ($P = 0.018$), and 4 months ($P = 0.005$). The mean attachment level values at 2 and 4 months post-therapy improved significantly in the M+A group compared to the placebo group ($P = 0.001$). Treatment with M+A resulted in a significant mean reduction in probing depth at 2 and 4 months compared to baseline values ($P = 0.001$). The M+A group showed a significant reduction of sites with high levels of *Pg* ($P = 0.001$) at 2 months compared with baseline values, and there was a significant reduction of sites with *Pg* and *Pi* in the M+A group compared with the placebo group. The results showed that a combined M+A treatment as the only therapy changes the proportion of some subgingival microorganisms and allows a significant improvement in clinical conditions.
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The goal of periodontal therapy is to preserve dentition by arresting, slowing down, or reversing periodontal destruction and to prevent the recurrence of the disease.[1] Successful periodontal treatment is dependent on a significant reduction of the commensal flora and of exogenous periodontopathogens in the subgingival microflora. Scaling and root planing reduce the proportion of pathogenic microorganisms, and their reduction is correlated to an improvement of clinical parameters.[2–3] Chemical agents have been used to obtain and maintain periodontal health as an adjunct to mechanical treatment to enhance its effect, and sometimes to facilitate or to replace mechanical treatment.[4] Metronidazole is a strong bactericidal agent that acts against strictly anaerobic bacteria by inhibiting the replication and the transcription of the bac-

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terial DNA.[5] Metronidazole is well established as a valuable chemotherapeutic agent, and it has been shown that systemic administration of metronidazole exerts beneficial effects on the clinical signs of chronic periodontal disease.[6–12]

Loesche et al.[7] demonstrated significant clinical improvement when metronidazole was used as the only treatment and also when it was combined with subgingival instrumentation in a pilot study involving 5 patients not given oral hygiene instruction. Metronidazole plus amoxicillin has been used successfully in the treatment of advanced periodontitis, especially with *A. actinomycemcomitans*-associated infections.[13,14] In most studies, metronidazole plus amoxicillin has demonstrated a positive effect on periodontitis in the presence of good oral hygiene and/or subgingival debridement. There is no information on the effect of the combination of metronidazole plus amoxicillin in the absence of mechanical therapy.

The present report describes the short-term results of research to determine the effects of metronidazole plus amoxicillin (M+A) on adult periodontitis. The study was designed to establish whether a 7-day course of metronidazole plus amoxicillin (M+A) as the only treatment would have any effect in arresting the progression of moderate to advanced adult periodontitis. The null hypothesis tested was that the clinical parameters in patients with adult progressive periodontitis treated with M+A as the only therapy are not statistically different from patients treated with a placebo.

MATERIALS AND METHODS

Sixty patients (18 men and 42 women; 36 to 68 years old; mean age 43.6 ± 8 years) were selected from the pool of recall patients attending a Public Dental Service and the Periodontics Clinic, Dental School of the University of Chile, Santiago. All patients had at least 14 treatable natural teeth, 4 of which were molars, at least 4 pockets ≥4 mm and 6 sites with attachment loss >3 mm and radiographic evidence of moderate to advanced destructive periodontal disease.

Exclusion criteria included medical conditions requiring premedication with antibiotics for periodontal probing; administration of medications such as antibiotics, steroids, or non-steroidal anti-inflammatory drugs within the last 6 months; or systemic diseases that might affect periodontal disease activity. Prior to the start of the study each subject received a supragingival prophylaxis to remove gross calculus thus allowing access for probing. The patients were monitored over a period of approximately 8 months, until at least 2 sites showed activity determined by ≥2 mm attachment lost. All patients were informed that the aim of the study was to investigate the effect of a new antibiotic combination to treat periodontal disease. All patients signed an informed consent before participating in the study, and the protocol was approved by the Review Committee for Ethical Norms of the Faculty of Dentistry, University of Chile. The protocol research stated that if a patient showed one or more teeth exhibiting attachment loss in 2 successive or alternate visits or a periodontal abscess during the study period, the tooth or teeth would then be treated with root planing and excluded from the study.

Clinical Measurements

The following variables were determined at the beginning of the study and at 2 and 4 months. 1) Oral hygiene status. The presence of continuous plaque at the cervical portion of the buccal, mesial, lingual, and distal surfaces of each tooth was recorded. 2) Gingival inflammation. Dichotomous measures of bleeding, 30 seconds after probing to the bottom of the periodontal pocket, was determined at the same 6 sites on each tooth on which probing pocket was measured. 3) Probing depth and relative attachment level measurements. Measurements were made at 6 sites on each tooth at the mesiobuccal, buccal, distobuccal, distolingual, lingual, and mesiolingual positions. Two models of the Florida Probe† were used. The Florida Disk Probe was used for relative attachment level recordings and the Pocket Depth Probe was used to make probing depth recordings. The probe was used with a titanium tip of 0.4 mm diameter; the measurement interval was 0.2 mm, and the applied load was 20 g.

All teeth, with the exception of third molars, were scored for probing depth and clinical attachment level. Only vertical probing depth and attachment level measures were made in molars with furcation involvement. A second attachment level measurement and probing depth were taken within 7 days of the first measurement so that a pair of attachment level measurements and probing depths were made every 2 months. The mean of the pair of measurements were used in all the analyses described below. Two calibrated examiners monitored the patients, but all clinical measurements taken on each individual, were made by the same examiner at each time period. Patients were monitored over a period of approximately 8 months, and all clinical measurements were taken at 2-month intervals until at least 2 sites showed ≥2 mm clinical attachment loss, or until the occurrence of a periodontal abscess.

Forty-six of the 60 subjects exhibited either at least 2 sites with ≥2 mm attachment loss, or the occurrence of periodontal abscess during one of the 2-month periods. They were randomly assigned to receive either 21 tablets of metronidazole 250 mg plus amoxicillin 500 mg or 21 tablets of a placebo and directed to take 1 tablet every 8 hours for the following 7 days. Metronidazole plus amoxicillin tablets and placebo tablets had the same appearance and were packed in identical containers. Neither exam-

†Florida Probe Corporation, Gainesville, Fl.

iners nor patients was aware of the identity of the tablets. Patients were advised not to consume alcohol during the week they were to take the medication. Smokers were randomly assigned to both groups. At 2 months and 4 months after taking the medication or placebo, clinical examinations were again recorded. At 2 months, patients were asked to return the pill containers. Compliance was assessed by counting the remaining tablets. Throughout the study, no effort was made to change the oral hygiene habits of patients, and they received no additional periodontal therapy during the 4-month period.

Microbiological Procedures

Identification of *Actinobacillus actinomycetemcomitans, Porphyromonas gingivalis,* and *Prevotella intermedia* was assessed utilizing DNA probe technology in both groups at day 0 and 2 months after therapy. Two active sites were selected for the microbiological sampling from each patient. Loosely adherent supragingival plaque located in direct proximity to the sample sites was carefully removed using a scaler and cotton gauze. A medium size, sterile, absorbent paper point was gently inserted into the apical extent of the periodontal pocket. After 10 seconds, the paper point was removed and placed in a sterile Eppendorf vial and sent to a commercial firm[‡] where it was analyzed for the presence of *A. actinomycetemcomitans, P. gingivalis,* and *P. intermedia.* For *A. actinomycetemcomitans,* a cloned probe was used, whereas for the other organisms whole genomic probes were applied. The laboratory results were reported in numerical values between $<10^3$ and $>10^5$ cells. Interpretation of the results was as follows: when the pathogen level was less than 0.1% of total or was less than 10^3 cells, the score was negative; a range from 0.1% to 0.9% of total or 10^3 cells was scored as low; range from 0.1% to 9.9% of total or 10^4 cells was scored as moderate; and when pathogen was greater than 10^5 cells was scored as high.

Statistical Analyses

Differences of clinical measurements between experimental and control groups at baseline was assessed using the *t* test. The variables evaluated were changes in mean attachment level in patients and the percentage of sites that gained or lost ≥2 mm attachment level. Change in attachment level was computed for each site, averaged within a patient and then averaged across individuals in experimental and control groups. Changes in probing depth at pre- and post-therapy, percent of sites bleeding on probing, and plaque accumulation were tested in the same way. The significance (alpha = 0.05) of differences between the 2 groups was assessed using the Mann-Whitney test. The significance of differences in attachment level, probing depth, bleeding on probing, and plaque accumulation at baseline and at 2- and 4-months post-therapy within each group was assessed using the ANOVA test for univariate repeated measures. A statistical package[§] was used in all data analyses. In the analysis of the microbiological results, sites positive for the pathogens were expressed as percentage of the total sites sampled in the subject groups. The nonparametric test of Fleiss was used to determine significant differences in proportions of sites with pathogens.

Table 1. Clinical Characteristics (mean ± SD) of M + A and Placebo Patients Prior to Treatment

	M + A (n = 23)	Placebo (n = 21)
Age (years)	43.7 ± 7.5	43.5 ± 8.8
N missing teeth	5.95	4.14
% males	14.28	13.04
Mean probing depth (mm)	2.74 ± 0.77	2.54 ± 0.58
Mean attachment level (mm)	3.4 ± 1.3	3.2 ± 1.1
% active sites	3.21 ± 1.63	3.07 ± 1.4
% sites with		
Plaque	62.39	62.71
Bleeding on probing	32.95 ± 14.29	28.95 ± 13.52
Probing depth <4	85.65 ± 10.4	88.95 ± 9.37
Probing depth 4–6 mm	12.09 ± 9	10.1 ± 8.94
Probing depth >6 mm	2.73 ± 2.73	1.88 ± 1.78
% smoking subjects		
5–10 cigarettes/day	30.43	28.57
>10 cigarettes/day	13.04	14.28

RESULTS

At 2 months, assessment of compliance of tablets ingestion was determined by counting the remaining pills in the returned containers. All patients reported they had taken the medication as indicated, and no complaints relative to medication were reported. Two patients in the placebo group were dropped at 2 months because they had taken an antibiotic for medical reasons; these patients were excluded from data analysis. Thus, the M+A group consisted of 23 subjects and the placebo group of 21 subjects.

Clinical Results

The immediate pretreatment clinical features of the subjects treated with M+A and subjects with placebo are presented in Table 1. No age or gender differences existed between the 2 groups. There were no significant differences in the clinical parameters at baselines between the 2 groups. Table 2 presents the changes in the mean of clinical parameters in the M+A group and in the placebo group at 2- and 4-months post-therapy.

There was a slight and nonsignificant decrease of surfaces with plaque in both groups at 2 months, and a slight increase at 4 months post-therapy. When the values at 2- and 4-months were compared to the baseline values within each group, there were more significant changes within

[‡]OmniGene, Cambridge, MA.

[§]Systat 7.0, SPSS, Chicago, IL.

Table 2. Change in Clinical Parameters in the M + A and Placebo Groups at 2 and 4 Months Post-Therapy

Clinical Parameter (% of sites)	M + A Group (n = 23)			Placebo group (n = 21)		
	Baseline	2 Months	4 Months	Baseline	2 Months	4 Months
With plaque	62.4 ± 20.2	59.9 ± 16	63 ± 17.7	62.7 ± 17	59.5 ± 17.8	65.3 ± 18.3
Bleeding on probing‡‡	32.9 ± 14.3	20.4 ± 11.6*	20.3 ± 8.4†	28.9 ± 13.5	31.2 ± 15.9*	34.2 ± 17†
Active sites‡‡	3.1 ± 1.63	1.18 ± 1.21‡	0.98 ± 0.84§	3.07 ± 1.4	3.14 ± 2.21‡	3.84 ± 1.79§
Gaining attachment level	0	1.4 ± 1.72‖	2.23 ± 1.91¶	0	0.59 ± 1.9‖	0.8 ± 1.15¶
Mean attachment gain (mm)	0	0.27 ± 0.24#	0.29 ± 0.19	0	0.096 ± 0.09#	0.12 ± 0.19
Mean attachment loss (mm)	0	0.18 ± 0.09**	0.10 ± 0.86††	0	0.22 ± 0.18**	0.32 ± 0.31††
Mean probing depth (mm)‡‡	2.74 ± 0.77	2.54 ± 0.79	2.50 ± 0.69	2.45 ± 0.58	2.60 ± 0.64	2.62 ± 0.66

*$P = 0.018$.
†$P = 0.005$.
‡$P = 0.001$.
§$P = 0.001$.
‖$P = 0.05$.
¶$P = 0.005$.
#$P = 0.001$.
**$P = 0.017$.
††0.006.
‡‡M + A group: Baseline values versus 2 and 4 months values ($P = 0.001$).

the M+A group than within the placebo group. The percentage of bleeding on probing sites in the M+A group decreased significantly from baseline to 2 months ($P = 0.001$) and at 4 months, while the percentage of bleeding sites in the placebo group increased at 2 and 4 months. However, the differences compared to baseline data were not significant in the placebo group. The differences of bleeding sites between M+A group and the placebo group were significant at 2 months ($P = 0.018$) and at 4 months ($P = 0.005$). At 2 months, an average gain in attachment was found in 21 patients (91%) of the M+A group and in 9 patients (43%) in the placebo group, and the difference was significant ($P = 0.002$). An average loss in attachment was found in 2 patients in the M+A group and in 11 patients in the placebo group. At 4 months, 21 patients (91.3%) of the M+A group while only 4 patients (19%) in the placebo group exhibited an average gain in attachment ($P = 0.001$). An average loss in attachment was found in 2 patients in the M+A group and in 14 patients in the placebo group. One patient in the placebo group showed no change in attachment level at 2 months, and 3 patients in the same group exhibited no change at 4 months.

The proportion of sites losing ≥2 mm attachment level, or active, sites in the M+A group decreased significantly at 2 and 4 months ($P = 0.001$) compared to baseline values. Active sites in the placebo group increased from baseline at 2 and 4 months, but not significantly ($P = 0.839$). The percentage difference of active sites between the M+A group and the placebo group was highly significant at 2 months ($P = 0.001$) and at 4 months ($P = 0.001$) post-therapy. The percentage of sites gaining attachment level was significantly higher in the M+A group compared to the placebo group at 2 ($P = 0.05$) and 4 months ($P = 0.005$).

The mean attachment gain in relation to baseline for patients treated with M+A was significant at the 2- ($P = 0.006$) and 4-month examinations ($P = 0.001$). The mean attachment gain for patients in the placebo group was not significant at either the 2- ($P = 0.301$) or 4-month examination ($P = 0.618$).

The mean attachment level values at 2- and 4-months post-therapy for patients in the M+A group improved significantly ($P = 0.001$) compared to the mean attachment levels for patients in the placebo group. The mean attachment loss value was significantly lower for the M+A group than for the placebo group at 2- ($P = 0.017$) and at 4-month examinations ($P = 0.006$). Treatment with M+A resulted in a significant mean reduction in probing depth of 0.20 mm at 2 months ($P = 0.001$) and of 0.24 mm at 4 months, while in the placebo group there was a mean increase in probing depth of 0.15 mm at 2 months, and of 0.17 mm at 4 months, which were not significant ($P = 0.175$) when compared with baseline values.

Microbiological Results

At day 0, microbiological data of 44 samples from 22 patients from the M+A group, and 36 samples from 18 patients from the placebo group were available. At 2 months, microbiological data were available from 36 samples of 18 patients of the M+A group, and from 30 samples of 15 patients from the placebo group. The microbiological data for all patients were not available because some samples were lost in the mail.

Table 3 shows the occurrence of *A. actinomycetemcomitans, P. gingivalis,* and *P. intermedia* in patients before and after treatment with M+A, and Table 4 shows the occurrence of the same pathogens in the placebo group. There were no significant differences between the occurrence of the *P. gingivalis* and *P. intermedia* in the M+A group compared with the placebo group at day 0.

The percentage of sites with *A. actinomycetemcomitans*

Table 3. Occurrence of *A. actinomycetemcomitans, P. gingivalis,* and *P. intermedia* in Active Sites of Patients M + A Before and After Therapy

Level of Detection	% Sites Before Therapy (n = 44)			% Sites After Therapy (n = 38)		
	Aa	*Pg*	*Pi*	*Aa*	*Pg*	*Pi*
High	0.00	59.09*	2.27	0.00	18.42*	0.00
Moderate	0.00	2.27†	18.10	0.00	26.31†	0.00
Low	6.81	11.36	25.00	5.26	15.78	23.68
Total	6.81	72.72	45.45	5.26	60.52	23.68

*$P = 0.001$.
†$P = 0.004$.

Table 4. Occurence of *A. actinomycetemcomitans, P. gingivalis,* and *P. intermedia* in Active Sites of Placebo Patients Before and After Therapy

Level of Detection	% Sites Before Therapy (n = 36)			% Sites After Therapy (n = 30)		
	Aa	*Pg*	*Pi*	*Aa*	*Pg*	*Pi*
High	0.00	50.00	0.00	0.00	40.00	0.00
Moderate	2.77	8.33	13.88	0.00	23.33	10.00
Low	0.00	19.44	36.11*	3.33	23.33	63.33*
Total	2.77	77.77	50.00	3.33	86.66	70.00

*$P = 0.050$.

was much lower than sites harboring *P. gingivalis* and *P. intermedia* in both groups and was significantly higher in the M+A group compared with the placebo group ($P = 0.004$) at day 0 (Tables 3 and 4). Patients treated with M+A showed a significant decrease of sites with high level of *P. gingivalis* ($P = 0.001$) and a significant increase of sites with moderate levels of *P. gingivalis* at 2 months, compared with baseline values. *P. intermedia* was not detected at high or at moderate levels in any site 2 months after M+A therapy, and the occurrence of this bacterium decreased almost 50% compared to day 0, but the difference was not significant. The occurrence of the 3 pathogens, not considering the level of detection, decreased in the M+A group, but the differences were not significant compared with day 0 values (Table 4). The occurrence of the 3 pathogens in the placebo group increased from day 0 to 2 months, but the only significant difference was the percentage of sites exhibiting low levels of *P. intermedia* ($P = 0.050$). When the prevalence of the 3 pathogens in the M+A group 2 months after therapy was compared with the prevalence of the same pathogens in the placebo group, a significant decrease in the percentage of sites with *P. gingivalis* ($P = 0.035$) and *P. intermedia* ($P = 0.001$) was found in subjects treated with M+A.

DISCUSSION

The results of the present study showed that systemic administration of M+A in moderate to advanced progressive adult periodontitis, as the sole treatment, reduced the percentage of bleeding sites, the proportion of active sites, and the probing depths. Two and 4 months after the M+A therapy there was also an increase in the proportion of sites gaining attachment level, and a significant improvement in the mean of clinical attachment level (Table 2). The improvement of the clinical parameters was associated with a significant decrease in percentages of sites harboring high levels of *P. gingivalis* and with the elimination of sites with high and with moderate levels of *P. intermedia* (Table 3).

An important characteristic of the current study was that all patients had shown disease progression in the 2 months prior to antibiotic therapy. Periodontal tissue destruction occurs in periodontitis as a dynamic condition of disease exacerbation and remission, as well as inactivity for unknown periods of time.[15–18] For this reason it seems essential in clinical trials to discriminate between situations in which an active aggressive infection is being treated from situations in which the treatment is only lowering the risk of disease in the near future.[19] The patients in the current study were recruited only after at least 2 sites losing ≥2 mm attachment level were diagnosed. This criterion has been employed in a number of other clinical studies.[20–26] In addition, every possible precaution to minimize the changes due to experimental error (i.e., replicate measurements, the same operator, and longitudinal measurements) were taken.

Successful periodontal treatment is dependent on changes in the microbiological composition of the subgingival microflora. Therefore, the goal of the treatment of periodontal disease is to eliminate or decrease the proportion of pathogens to a level manageable by the host. Systemic periodontal antimicrobial therapy is based on the premise that specific microorganisms cause destructive periodontal disease and that the antimicrobial agent in the periodontal pocket must reach the concentrations necessary to eliminate the pathogens.[27] It has also been

suggested that in the treatment and control of periodontal disease, the selective elimination of pathogenic microorganisms, rather than the subgingival microbiota *in toto,* may be preferred.[28] Metronidazole has been shown to have a pronounced effect on the subgingival microbiota of periodontal lesions in humans.[6–11] Otherwise, amoxicillin appear to be very effective against most periodontal pathogens[29–31] and exhibits high antimicrobial activity at levels that occur in gingival crevicular fluid.[32] The combination of M+A has been shown as an effective adjunctive therapy for the elimination of *A. actinomycetemcomitans*[13,33] and it would be expected to target a broad spectrum of periodontal organisms with metronidazole inhibiting the anaerobes, and amoxicillin inhibiting the facultative and aerobic bacteria.[34] In most clinical studies, systemic metronidazole has been shown to augment the clinical effect of mechanical periodontal treatment in periodontitis patients. However, metronidazole therapy without concomitant scaling and root debridement provides very short-lived clinical and microbial benefits.[8,35] According to Loesche et al.,[36] metronidazole is most effective when given to patients with high proportions of spirochetes in their plaque, and when combined with mechanical debridement. The equivocal results obtained in some studies[12,37–39] more probably reflect the inadequacies of metronidazole when used either in the absence of an anaerobic infection, or in advanced clinical disease.[36] The significant improvement of clinical parameters in patients treated with M+A as the sole treatment may be ascribed to the synergistic effect between amoxicillin and metronidazole against periodontal pathogens. The combined effect of M+A may overcome the low efficacy of metronidazole when used either in the absence of a predominant anaerobic infection, or in advanced periodontal disease. The favorable effects of M+A on moderate to advanced adult periodontitis patients who had not received any additional periodontal therapy during the 4-month follow up period provides a rationale for the use of this antibiotic combination as an adjunctive therapy to root planing.

The significant improvement in all the clinical parameters in patients treated with M+A is surprising, considering that there was no additional periodontal treatment during the 4-months, and no efforts were made to instruct the patients in supragingival plaque control. All the patients included in the present study had poor oral hygiene as shown by the high percentage of surfaces with dental plaque, which remained almost unchanged at 2 and 4 months. Some studies have suggested that failing to obtain an appropriate level of plaque control after therapy may reduce the success of therapy.[40,41] There is a significant discrepancy in the literature regarding the effect of dental plaque removal and of supragingival plaque control on the subgingival microflora.[42] The results of the present study suggest that supragingival plaque levels may have no influence on the short-term results of the therapy if the antimicrobial therapy eliminates or reduces the pathogenic microorganisms of the subgingival microbiota. The possible changes in subgingival microflora ascribed to the effect of M+A are apparently independent of the presence of supragingival plaque. However, the possibility that the antibiotic levels in saliva may have had some effect on the microbial composition of supragingival plaque cannot be excluded, since metronidazole is found in saliva at levels which are much higher than that required for inhibition of *Treponema, Bacteroides* and other anaerobic organisms.[43]

Short-term effects of periodontal treatment have traditionally been measured as a reduction of gingival inflammation and probing depths and as a gain in clinical attachment level. It has been found that patients differ in their response to periodontal therapy as assessed by the clinical attachment level post-therapy. In the present study, 2 patients of the M+A group showed loss of attachment level 2 months after therapy. One of these patients continued losing attachment at 4 months, and the other patient gained attachment at 4 months. A third patient exhibited gain of attachment at 2 months and loss of attachment at 4 months. Haffajee et al.[43] found that poor response to therapy was associated with the lack of decrease in gingival inflammation. The relationship between persistence of gingival inflammation and poor response to therapy was not found in the present study since all 3 patients who showed a poor response to therapy exhibited a significant decrease in the percentage of sites bleeding on probing after treatment. A poor response to therapy may suggest that a significant proportion of the subgingival microflora of those patients remained unaffected following M+A administration, or that the microbial changes in the subgingival microflora were of too short a duration to produce a favorable effect on the clinical attachment level 2 months after therapy. A recent study of Flemmig et al.[44] on the effect of systemic administration of M+A as an adjunct to scaling showed that *P. gingivalis* persisted in subgingival plaque after therapy in 13 out of 15 patients who initially harbored this pathogen. The results of Flemmig et al.[44] and the results of the present research agree with many other studies that showed that the elimination or the reduction of certain periodontal pathogens from the subgingival plaque is a transient effect of the mechanical periodontal therapy, and of the antibiotic therapy.

The most significant effect of the M+A therapy in the present study was found at 2 months. At the 4-month examination there was an additional slight and nonsignificant improvement in the clinical parameters. These results may suggest that, after 4 months, no significant additional improvement could be obtained by using M+A as the only periodontal therapy. This transient effect of M+A on subgingival microflora is possibly due to a ma-

jor effect of the antibiotics on bacteria that are in suspension or not adhering to the biofilm subgingival dental plaque.[46]

The results of the present short-term, double-blind study showed that a 1-week course of systemic metronidazole plus amoxicillin, as the sole therapy in patients with moderate to advanced adult periodontitis, alters the composition of the subgingival plaque allowing a significant improvement in the clinical conditions. Further long-term studies aimed at determining if the M+A therapy, given in intermittent periods, will lead to a permanent improvement of progressive periodontal disease have been undertaken.

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