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Reduction in Fever and Symptoms in Young Adults with Influenza A/Brazil/78 H1N1 Infection After Treatment with Aspirin or Amantadine

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During an outbreak of influenza A/Brazil/78 H1N1 infection, 47 volunteers with clinical and virological influenza of less than 2 days duration were treated in a randomized double-blind fashion for 5 days with 100 or 200 mg of amantadine daily or with 3.25 g of aspirin daily. The aspirin treatment group defervesced more rapidly (10.3 h versus 21.5 h and 23.6 h; $P < 0.01$), but by the second daily follow-up visit, both groups of amantadine recipients exhibited greater symptomatic improvement. Bothersome side effects resulted in discontinuation of therapy by 35% of the aspirin treatment group but only 3% of the amantadine treatment group ($P < 0.05$). Individuals who present to a physician during an influenza A epidemic with characteristic symptoms will experience symptomatic benefit from amantadine treatment, with negligible toxicity.

Studies performed over the past 15 years have confirmed the therapeutic benefit of amantadine when given early in the course of acute illness due to influenza A virus, both in shortening the duration of symptoms and in decreasing their severity (6, 10). Despite these reports, amantadine is not widely used by medical practitioners. Perhaps this is in part because many physicians are reluctant to initiate therapy without specific viral diagnosis and therefore must make a presumptive diagnosis of influenza on the basis of clinical findings. Second, clinicians may be concerned about the side effects of amantadine and may be convinced that other drugs, particularly aspirin, are just as effective in relieving symptoms and controlling fever.

During an outbreak of influenza A/Brazil/78 H1N1 infection in the winter of 1981, a controlled double-blind study was performed comparing the therapeutic advantages and disadvantages of amantadine and aspirin for the treatment of influenza. Furthermore, low and high doses of amantadine were compared to determine whether previously reported side effects (2, 4, 10) could be avoided while maintaining efficacy.

MATERIALS AND METHODS

Volunteers. This study was initiated when an outbreak of influenza A/Brazil/78 H1N1 was detected by our viral surveillance system (7). Volunteers were healthy college students, ages 17 to 20, recruited from the University Health Service. Before inclusion in the study, each volunteer was given a description of the objectives, procedures, risks, and benefits of participation, and written, informed consent was obtained. Volunteers remained in their usual campus living quarters.

Clinical evaluation. Volunteers who were admitted to the study had at least two of the following symptoms (of less than 48 h duration): headache, myalgia, cough, or feverishness. A complete history of the present illness was obtained from each student on admission, and follow-up interviews were performed 1, 2, 3, and 7 days thereafter. Specific complaints were grouped into the categories of upper respiratory (earache or obstruction, nasal discharge or obstruction, sore throat, hoarseness), lower respiratory (chest pain, cough), and systemic symptoms. The latter category included feverishness, chills, myalgias, malaise, headache, and anorexia. Each symptom was graded on a scale (0 indicating none; 1, mild; 2, moderate; 3, severe). A symptom was described as mild when there was a noticeable difference from normal and as severe when that symptom was as severe as previously experienced by that person.

In addition, patients were specifically questioned about symptoms of diarrhea, tinnitus, epigastric discomfort, anxiety, insomnia, or sleep disturbance to provide information about the frequency of medication side effects. Oral temperatures were recorded by the investigators at daily visits between 2:00 p.m. and 10:00 p.m., and volunteers were provided with thermometers and instructed to record their temperatures each night before sleep. A physical examination was not performed.

Virological studies. Nasal washings with veal infusion broth for viral isolation were obtained on admis-

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sion to the study and on each subsequent visit (3). They were maintained at 4°C overnight. Portions were inoculated onto Madin-Darby canine kidney cell (MDCK) monolayers, and the remaining specimens were frozen at −70°C (1). Monolayers were examined daily for evidence of cytopathic effect. All positive specimens were quantitated by titration of serial 10-fold dilutions of the frozen specimens in MDCK cells (12). Endpoints were calculated by using the Spearman-Karber method, and results were expressed as $\log_{10}$ 50% tissue culture infectious dose per milliliter. The first isolate from each volunteer was identified by hemagglutination inhibition with specific antisera, and antibody titers were performed on acute and 3-week convalescent sera by hemagglutination inhibition methods as well (5). By utilizing a plaque inhibition assay method, amantadine susceptibility of the influenza A/Brazil/78 H1N1 isolates from 1981 was determined simultaneously with that of isolates of influenza A/USSR/77 H1N1 from 1978 (8). Results were expressed as the concentration of amantadine required to produce 50% inhibition of plaque formation.

Study design. The purpose of this study was to compare the treatment of influenza A infection with 100 mg of amantadine daily, 200 mg of amantadine daily, or 3.25 g of aspirin daily in a prospective double-blind fashion. To achieve this goal, our research pharmacist prepared three sets of 20 large manila envelopes, each containing 5 small envelopes. Each of the small envelopes was labeled day 1, day 2, etc., representing a 5-day supply of medication.

One set of the large envelopes held five small envelopes, each of which contained two 100-mg amantadine tablets and 10 capsules containing inert filler. The second set of large envelopes held five small envelopes, each of which contained one 100-mg amantadine tablet, one identical-appearing placebo tablet, and 10 capsules containing inert filler. The final set of large manila envelopes held five small envelopes in which were two placebo amantadine tablets and 10 capsules, each containing 325 mg of acetylsalicylic acid. These were identical in appearance to the placebo capsules.

The 60 large envelopes were assigned numbers from a predetermined list which resulted in random distribution. When a volunteer was admitted to the study, he or she received the large envelope with the next number in sequence. On enrollment, the small envelope labeled "day 1" was removed from the large manila envelope. The two tablets and two of the capsules from that envelope were ingested at that time. That evening and for the next 4 days, students took two capsules at 4-h intervals, beginning at 8:00 a.m. and ending at midnight. On days 2 through 5, they also took one tablet at 8:00 a.m. and one at 8:00 p.m. from the appropriate day's envelope. Compliance was evaluated by daily questioning and by instructing students to return the packets on completion of the study. Participants were asked not to take additional aspirin or acetaminophen-containing medications during the study period. Differences between groups were analyzed by Student's t test.

RESULTS

Study population. The study population was composed of 48 college-aged individuals, with a similar number of males and females in each therapeutic group (Table 1). All had onset of the signs and symptoms of influenza infection within 48 h of enrollment; most presented on the day of onset of illness. Febrile individuals (oral temperature greater than 38°C on presentation) were evenly divided in all groups with a slight, but not significant, predominance in the groups receiving amantadine. In general, those who were initially febrile had been ill for less than 24 h at the time of presentation. An equal number of participants in each group initially had unusually severe illness, as manifested by a total symptom score greater than 35. In addition, the duration of illness in the afebrile or febrile subgroups of each treatment group was similar. Of the 48 subjects, 47 shed influenza A/Brazil/78 H1N1 virus on at least one occasion. Furthermore, the number of subjects with detectable virus in the first nasal washing and the mean titer of virus in that specimen were similar in all three groups.

TABLE 1. Study population[a]

Characteristic	No. of volunteers in following treatment group:		
	Aspirin	100 mg of amantadine	200 mg of amantadine
Total in group	17	16	14
Males/females	8/9	9/7	6/8
Febrile on presentation (temp of 38°C)	9 (53)%	12 (73%)	8 (57%)
Initial symptom score greater than 35	3 (18%)	3 (19%)	3 (21%)
Shedding influenza A virus on day 1	14	15	10

[a] The mean influenza virus titer ($\log_{10}$ 50% tissue culture infectious dose per milliliter) on day 1 of those volunteers shedding virus was 2.6 for the aspirin group, 1.9 for the 100-mg amantadine group, and 2.0 for the 200-mg amantadine group.

Clinical syndrome. Headache, myalgias, cough, and feverishness were uniform complaints. More than three-fourths of the participants experienced anorexia, which usually persisted for the duration of illness, but only 14 (29%) had nausea or vomiting. Three students had mild diarrhea, and this occurred at least 24 h after admission to the study.

There were generalizations that could be made about the response of the study population to therapy. Although all but four volunteers were no worse or somewhat better at the first follow-up visit, improvement was slight and did

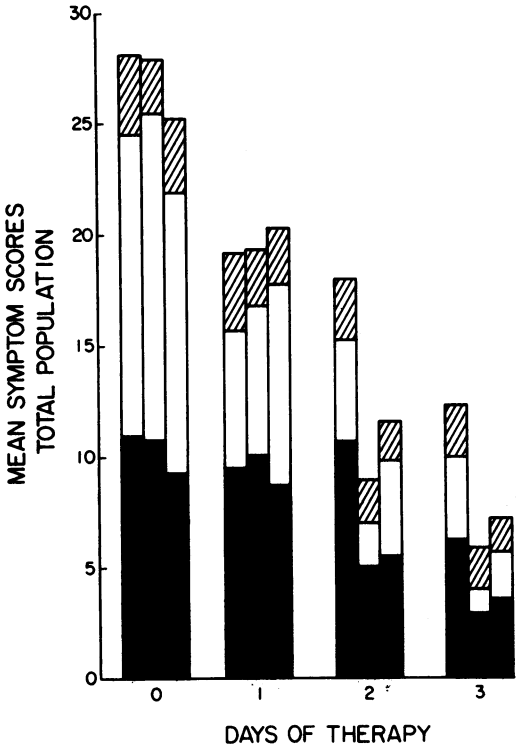

FIG. 1. Mean symptom scores by day according to type of symptoms. In each set of three bars, the aspirin group is on the left, 100 mg of amantadine is in the middle, and 200 mg of amantadine is on the right. Symbols: ▨, lower respiratory symptom score; □, systemic symptom score; ■, upper respiratory symptom score.

not differ among treatment groups. It was not until the second and particularly the third daily visit that obvious symptomatic improvement was seen, and by that time the majority had returned to classes. Cough was a persistent but not particularly bothersome complaint, and no difference could be detected in its severity among the treatment groups.

Response of fever. Considering only the 29 subjects (62%) who had an initial temperature greater than 38°C (Table 1), the aspirin-treated group defervesced most rapidly. They achieved a temperature of less than 38°C in a mean time of 10.3 h, compared to 21.5 h for the group taking 100 mg of amantadine daily (Student's t test; $P < 0.01$) and 23.6 h for the group taking 200 mg of amantadine daily.

Symptomatic response. Twenty-four hours after presentation, there were no significant differences in symptom scores among the three treatment groups. It was not until the 2- and 3-day follow-up visits that differences were noted. When mean daily symptom scores were tabulated, the volunteers receiving 100 mg of amantadine daily had significantly lower values at 48 and 72 h than did the volunteers receiving aspirin ($P < 0.01$; Fig. 1). Although the group who received 200 mg of amantadine daily had substantially lower overall symptom scores than the aspirin treatment group, this difference did not achieve statistical significance ($0.05 < P < 0.1$; Fig. 1). When analysis was limited to the category of "systemic symptoms," results again followed this pattern. The 100- and 200-mg-amantadine groups did better than the aspirin group, but only for the 100-mg group were the differences significant. In the category of "upper respiratory" symptoms, both amantadine-treated groups experienced greater symptomatic improvement at the 48-h follow-up visit than the aspirin-treated individuals ($P < 0.05$; Fig. 1). Analysis of return to classes was made difficult by weekends intervening in the study and by different class schedules of participants, but there was no obvious difference in the time in which students returned to class.

If the subgroup of subjects whose initial temperature was greater than 38°C was analyzed separately, significant differences could be seen, favoring the group treated with 100 mg of amantadine daily over the aspirin-treated group in both overall symptom scores and "systemic" symptoms at 48 and 72 h ($P < 0.05$). Once again, similar but not significant differences were seen between the groups treated with 200 mg of amantadine daily and aspirin.

Virological data. There were no significant differences among the three groups with respect to frequency of virus shedding on any of the study days (Tables 1 and 2). In addition, there were no significant differences in mean daily virus titers among the three treatment groups on the day of enrollment or any subsequent day (Tables 1 and 2). Furthermore, little decrease in virus titer was detected, despite a substantial decrease in symptom scores. The only difference between the groups with respect to virus shedding was seen in the high-dose amantadine group. Analysis of the number of days of virus shedding versus the number of tested days of shedding after entry revealed that, in the high-dose amantadine group, only 22 of 42 culture days were positive, versus 40 of 51 in placebo recipients ($P < 0.02$). The number of days positive in the low-dose amantadine group was 33 of 48 (P not significant). Of the 47 subjects, 38 had a fourfold or greater rise in antibody titer between acute and convalescent specimens. The 1981 influenza A/Brazil/78 H1N1 isolates were uniformly sensitive to amantadine by plaque inhibition assay. The 50% inhibitory concentration of amantadine for three isolates obtained on day 1 was less than 0.125 μg/ml, consistent with

TABLE 2. Effect of amantadine treatment on influenza virus recovery and titers

Treatment group	No. of volunteers shedding virus on day:			Virus titer[a] on day:		
	2	3	4	2	3	4
Aspirin	13	15	12	1.9	1.7	1.6
100 mg of amantadine	11	12	10	1.2	1.5	1.8
200 mg of amantadine	8	9	5	1.4	1.3	1.1

[a] Mean virus titer for those volunteers shedding virus on $\log_{10}$ 50% tissue culture infectious dose per milliliter.

results obtained concurrently by using three day-1 isolates of influenza A H1N1 from 1978.

Side effects. A number of volunteers in all groups experienced a symptomatic complaint on at least one occasion that they attributed to the medication. In the aspirin treatment group, the subjects took all tablets, but six did not take all prescribed capsules. All subjects took all medications the first 3 days of the study. Six patients also had at least one episode of insomnia, nausea, or tinnitus. In the low-dose amantadine groups, six patients complained of dizziness, loss of concentration, or insomnia, and all but one completed the full course of tablets and placebo capsules. Four patients who received 200 mg of amantadine daily had insomnia, but all were completely compliant and did not feel that these symptoms significantly impaired daily activities. Significantly more volunteers discontinued aspirin treatment than amantadine treatment because of side effects ($P < 0.05$).

DISCUSSION

College-aged volunteers with naturally acquired Influenza A/Brazil/78 H1N1 infection who received a total daily dose of 3.25 g of aspirin defervesced more rapidly than volunteers who received either 100 or 200 mg of amantadine daily. However, by the end of the second day of treatment, a significant therapeutic benefit of amantadine compared to aspirin could be demonstrated in the subgroup consisting of those who were febrile at presentation and in the entire patient population. The benefit occurred as measured by systemic symptom scores as well as upper respiratory symptom scores, although neither amantadine nor aspirin had a superior effect on cough. These findings are consistent with a previous study from this institution (16) which demonstrated that amantadine therapy shortened the duration of illness in comparison to placebo and with other published studies (11, 15, 17). In this study (amantadine versus aspirin) and in our previous study (amantadine versus placebo), amantadine shortened the duration of illness by 24 h or roughly 33% of the illness duration after treatment (16). Considering the different year and different subjects, this is an interesting consistency. Furthermore, it is surprising that amantadine proved to be superior in reducing symptoms, since lysis of fever occurred earlier in aspirin recipients. The present study also suggests that a single 100-mg tablet of amantadine daily has therapeutic efficacy equal to that of a higher dose. It is noteworthy that, because of the study design, some individuals may have had 36 h elapse between amantadine doses and yet a therapeutic response still occurred.

In the assessment of a therapeutic regimen for self-limited disease, both the beneficial and adverse effects contribute to the acceptance of the regimen. About one-third of the amantadine-treated groups had minimal central nervous system side effects described as loss of concentration or insomnia. Only one volunteer found these side effects troublesome enough to discontinue therapy. Side effects were more severe in the aspirin-treated group (particularly nausea and tinnitus), leading to the failure of more than one-third of this group to complete treatment. Because participants were informed at the outset of the study that the capsules contained either aspirin or placebo, we presume that the aspirin-treated group stopped taking capsules because they attributed the symptoms they were experiencing to aspirin. Findings of other investigators with respect to amantadine toxicity are noteworthy. Van Voris and co-workers (16) found that 33% of amantadine recipients experienced minor central nervous system side effects by day 5 of therapy, but less than 5% failed to take all medications. Hayden et al. (9) concluded that 200 mg of amantadine daily was very well tolerated by healthy volunteers over a short treatment period, although 300 mg daily lead to unacceptable side effects. Bryson et al. (2) also described central nervous system side effects in 33% of a large group of college students treated with 200 mg of amantadine daily for 4 weeks. Ten percent discontinued therapy before the study was completed. No assessment of the side effects of aspirin in influenza patients has been made previously. The present study suggests that aspirin is of benefit on day 1, but that, beyond the first 24 h of treatment, not only does amantadine have a more beneficial effect on influenza symptoms but it also has less bothersome side effects. Although side effects due to amantadine in this population were minimal, until it is certain that amantadine can be tolerated by the elderly and by other groups in whom dexterity is important, use must be individualized.

The effect of amantadine on frequency or titer of virus shedding has not been entirely consistent in previous studies. Togo et al. (15) could not detect an effect of amantadine treatment on the number of patients who shed influenza virus in sequential nasal washes over a 5-day treatment period, although quantitation was not done. Although Knight et al. (11) could demonstrate a significant reduction in viral titers in amantadine-treated patients with influenza who had been ill for more than 2 days, there was no significant effect of amantadine treatment on those who had been ill for less than 48 h before institution of treatment. All of the patients in our study were ill for less than 48 h. Van Voris and co-workers (16) noted that amantadine was associated with significant reduction in the number of patients shedding influenza A/USSR/77 H1N1 after the second day of treatment. All of these subjects had been ill for less than 48 h as well. The majority of the volunteers in our study continued to shed virus 72 h after beginning therapy with either amantadine or aspirin (Table 2). In vitro sensitivity testing did not reveal amantadine resistance of the 1981 strain of influenza A/Brazil/78 H1N1 to account for these differences in the effect of drug on virus shedding. Although the percentage of patients shedding virus on day 4 was lower in the 200-mg amantadine group than the aspirin group, this trend did not achieve statistical significance ($P < 0.05$ by the chi-square test with Yates correction). Only when all 3 tested days were analyzed did there appear to be a difference in the high-dose amantadine group, a difference which did not occur in the low-dose group. This minimal effect on virus shedding is therefore consistent with other studies. Aspirin therapy did not appear to increase or prolong virus shedding (13). Finally, emergence of resistance to amantadine could not be tested due to failure of day-3 and -4 isolates to replicate serially after primary isolation in MDCK cells. These attempts were not made until after the isolates had been thawed and refrozen for purposes of titering. Presumably, these maneuvers had reduced the infectivity of the virus.

One other important point that this study confirmed was that individuals who present to a physician with acute onset of feverishness, cough, headache, myalgias, or anorexia but generally without vomiting or diarrhea during a known influenza outbreak can be assumed to have influenza without specific virological diagnosis. Of 48 students who presented with these symptoms, 47 had influenza A/Brazil/78 H1N1 virus detectable in nasal washings. Only 39 of these 47 volunteers with documented influenza grew virus from nose washes obtained on day 1, suggesting that clinical signs and symptoms are more accurate than a single specimen for virus isolation in making this diagnosis.

This study substantiates the therapeutic benefit of amantadine in naturally acquired influenza infection and demonstrates for the first time that a dose of 100 mg/day has equal therapeutic efficacy to one of 200 mg/day. Aspirin was shown to be particularly useful in producing defervescence on the first day of therapy, suggesting that it might be combined with amantadine in the first 24 h of influenza treatment. Side effects of aspirin preclude longer use. It has recently been demonstrated that Reyes syndrome occurs more commonly in children with influenza who are treated with aspirin, suggesting that acetaminophen might be more appropriate in this setting (14).

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