Cephacetrile: Clinical Evaluation in 27 Patients

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Cephacetrile, a new derivative of 7-aminocephalosporanic acid, was evaluated in 27 patients. Soft tissue infections due primarily to gram-positive cocci were treated in 16 patients; 12 had bacteriological and clinical cure, and 4 improved but the lesions resolved incompletely or cultures remained positive. Seven of eight patients with respiratory tract infections were cured, including three with pneumococcal pneumonia; the eighth proved to have a noninfectious process and failed to respond. Two patients with acute urinary tract infections due to *Escherichia coli* had prompt clinical and bacteriological improvement, but follow-up was incomplete. One patient with sepsis due to *Staphylococcus aureus* expired. Laboratory abnormalities observed during cephacetrile therapy included mild eosinophilia in four patients, thrombocytosis in nine, direct Coombs' test positivity in four, and an elevated serum glutamic pyruvic transaminase in eight patients. No evidence of nephrotoxicity was detected. Severe superinfection due to *Enterobacter* species was observed in one patient. Mean peak serum concentrations of cephacetrile were 22, 69, and 104 $\mu\text{g/ml}$ after 1 g intramuscularly, 1 g intravenously, and 1.5 g intravenously, respectively. Thus, in early studies cephacetrile was efficacious for selected bacterial infections, but determination of its comparative value within the cephalosporin group of antibiotics requires further clinical investigation.

The cephalosporin group of antibiotics has been widely accepted for the treatment of gram-positive coccal infections and selected gram-negative bacterial infections (15, 18). Although the two available parenterally administered cephalosporins, cephalothin and cephaloridine (Fig. 1), are of proven efficacy, new derivatives of 7-aminocephalosporanic acid are being evaluated in a quest to find parenteral agents with new or improved properties.

Cephacetrile, 7-cyanacetamido-cephalosporanic acid (Fig. 1), was evaluated in the present study in 27 patients with soft tissue, acute respiratory, acute genitourinary, or septicemic infections. The results of the evaluation form the basis of this report.

MATERIALS AND METHODS

Clinical aspects. Patients considered for inclusion in the study were males and nonpregnant females aged 18 years or more who were admitted to the Infectious Disease Service at the Ohio State University Hospital during the period September 1971 to January 1972. In each case, a presumptive diagnosis of bacterial infection due to a cephalosporin-susceptible organism was made on the basis of history, physical examina-

tion, and microscope examination of gram-stained material. Cephacetrile therapy was instituted only after the nature of the study and potential side effects of cephacetrile had been explained to the patients and written consent had been obtained.

Complete blood counts, serum glutamic pyruvic transaminase (SGPT), alkaline phosphatase, bilirubin, blood urea nitrogen (BUN), serum creatinine, fasting blood sugar, direct Coombs' test, and urinalysis were obtained prior to, at weekly intervals during, and at the end of cephacetrile therapy. Appropriate cultures were obtained initially to confirm the clinical impression, and follow-up cultures were obtained during and after therapy when possible.

Therapeutic cure was defined as complete clinical recovery with the absence of the initial pathogen in follow-up cultures; complete healing of wounds was accepted as evidence of bacteriological cure in instances where no lesions remained to be cultured. Certain patients were designated as "improved" if marked clinical response occurred but cultures remained positive or wound healed incompletely.

Laboratory aspects. The susceptibility to cephacetrile of bacterial strains isolated from patients was determined in vitro by twofold dilution techniques which have previously been described (10) and by diffusion tests performed with 60-, 30-, 25-, 10-, and 5- μg paper discs. Mueller-Hinton

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FIG. 1. Chemical structures of cephalothin sodium, cephaloridine, and cephacetrile sodium.

agar was used for diffusion tests except for *Diplococcus pneumoniae* and *Streptococcus pyogenes*, which were tested on sheep blood agar.

Drug concentrations in serum and urine were determined by the cylinder-plate method with *Staphylococcus aureus* ATCC 6538P as the test organism. Serum samples were diluted with a 1:1 mixture of normal human serum and 1% phosphate buffer (pH 6.0) to concentrations estimated to fall within the range of the standard curve. Urine samples for assay were diluted with sterile undiluted normal human urine. All diluents were pretested for presence of antimicrobial activity prior to use.

RESULTS

Soft tissue infections. Sixteen patients with soft tissue infections were initially treated with 4 to 9 g of cephacetrile per day (Table 1). Infections in 9 of 10 patients with cellulitis caused by a staphylococcus or a streptococcus, or both, were cured; one patient (no. 5) with cellulitis and abscess formation of the scalp had marked clinical improvement during therapy but relapsed 2 months later. One (no. 14) of two patients with negative cultures but suspected staphylococcal and/or streptococcal cellulitis had a clinical cure; the second patient (no. 3) had clinical improvement but incomplete healing because of the presence of a corn smut fungus (*Ustilago zeae*) in the lesion. Two of four patients with soft tissue

infections due to multiple organisms including a staphylococcus or streptococcus had therapeutic cure (no. 2 and 4); two had only clinical improvement. One of the latter two patients (no. 8) had ischemic gangrene and cellulitis of her right foot; cephacetrile therapy resulted in resolution of the cellulitis, but the gangrene progressed, necessitating a below-the-knee amputation. The other patient (no. 13) had massive cellulitis with abscess formation extending from the lower costal margin to the mid-thigh on the right side of his body. During cephacetrile therapy, there was complete clearing of the cellulitis; however, *Haemophilus aphrophilus* and *Bacteroides fragilis* were still present in the abscess drainage in later follow-up cultures.

Respiratory infections. Eight patients with respiratory tract infections were treated with cephacetrile (Table 2). Three patients with pneumonia due to *Diplococcus pneumoniae*, including one complicated by bacteremia, responded satisfactorily, as did three patients with suspected pneumococcal pneumonia. One of the latter three patients (no. 21) had a *Mycoplasma pneumoniae* complement-fixation titer of 1:512 on two occasions during his illness, suggesting preceding or concurrent *M. pneumoniae* infection. A patient (no. 22) with pneumonia due to *S. aureus* and concurrent *Proteus mirabilis* bacteremia became afebrile and had dramatic clinical improvement within 24 hr after institution of therapy. The single patient (no. 20) in the respiratory group who did not respond to cephacetrile therapy had vasculitis on muscle biopsy; the pulmonary infiltrates, initially interpreted as being due to infection, improved after cephacetrile was discontinued and steroid therapy was begun.

Miscellaneous infections. Two patients with acute pyelonephritis due to *Escherichia coli* were treated with cephacetrile (Table 3). Both patients had rapid clinical improvement and negative urine cultures after 3 days of therapy. However, neither patient returned for follow-up studies. One patient (no. 7) with apparent bacterial endocarditis and associated bacteremia due to an *S. aureus* strain which was sensitive to cephacetrile in vitro (Table 3) received the study drug for 6 days. Although significant clinical improvement occurred with clearing of embolic skin lesions and bacteremia, the patient expired apparently as a result of multiple cerebral emboli. Permission for an autopsy was not granted.

Patient tolerance. Nine of 21 patients who received cephacetrile intramuscularly had no complaints and no local reactions from the injections; 11 had mild discomfort with each injection which lasted less than 5 min, and 1 patient had more severe discomfort lasting up to 30 min. Of

TABLE 1. *Soft tissue infections treated with cephacetrile*

Patient no.	Age (years)	Sex	Disease	Pathogen(s)	MIC ($\mu\text{g/ml}$)	Daily dose (g)	Days of therapy	Clinical outcome
1	55	M	Cellulitis, scalp	<i>Staphylococcus aureus</i>	1.25	6 3	1 1 ^a	Cure
				<i>Streptococcus pyogenes</i>	ND ^b			
				<i>Streptococcus</i> sp., alpha-hemolytic	ND			
2	25	M	Cellulitis and abscess, arm	<i>S. faecalis</i>	1.25	6	5	Cure
				<i>Bacteroides melaninogenicus</i>	ND	3	1 ^a	
				<i>Klebsiella</i> sp.				
				<i>Enterobacter</i> sp.	10.0			
3	28	M	Cellulitis, finger	<i>Ustilago zeae</i>	ND	6	6 ^a	Im- prove- ment
				<i>S. pyogenes</i> ^c	—			
4	28	F	Mastitis	<i>S. faecalis</i>	0.62	6	3	Cure
				<i>S. aureus</i>	0.31	4 3	1 3 ^a	
5	36	M	Chronic cellulitis and abscesses, scalp	<i>Staphylococcus epidermidis</i>	0.16	6	2	Im- prove- ment
				<i>Peptococcus</i> sp.	ND	4	6 ^a	
6	23	F	Cellulitis, leg	<i>S. pyogenes</i>	0.31	6	4 ^a	Cure
8	62	F	Cellulitis, foot	<i>Escherichia coli</i>	10.0	4	8	Im- prove- ment
					ND			
9	20	F	Cellulitis, foot	<i>S. epidermidis</i>				
				<i>S. aureus</i>	1.25	6	8 ^a	Cure
				<i>Streptococcus</i> sp., beta-hemolytic	0.31			
10	42	M	Cellulitis, scalp	<i>S. pyogenes</i>	0.31	8	11	Cure
11	22	M	Cellulitis and abscess, sacrum	<i>S. aureus</i>	2.5	6	6 ^a	Cure
13	50	M	Massive cellulitis and abscess, right flank	<i>Haemophilus para-influenzae</i>	0.62	9	20	Im- prove- ment
				<i>H. aphrophilus</i>	>10.0			
				<i>B. fragilis</i>	ND			
				<i>B. melaninogenicus</i>	ND			
				<i>Streptococcus</i> sp., alpha-hemolytic	0.31			
				<i>Peptococcus</i> sp.	ND			
14	32	F	Cellulitis, leg	<i>S. aureus</i> and/or <i>S. pyogenes</i> ^c	—	6	4 ^a	Cure
18	23	M	Cellulitis and abscess, leg	<i>S. aureus</i>	1.25	9 4	2 7	Cure
23	28	M	Cellulitis and abscess, arm	<i>S. epidermidis</i>	0.62	6	11	Cure
26	38	M	Cellulitis, foot	<i>S. aureus</i>	1.25	6	7	Cure
27	26	M	Cellulitis, arm	<i>S. epidermidis</i>	0.31	4	5	Cure

^a Received follow-up therapy with cephalexin.^b Not done.^c Suspected pathogen.

TABLE 2. Respiratory tract infections treated with cephacetrile

Patient no.	Age (years)	Sex	Disease	Pathogen(s)	MIC ($\mu\text{g/ml}$)	Daily dose(g)	Days of therapy	Clinical outcome
12	27	F	Pneumonia	<i>Diplococcus pneumoniae</i>	0.16	6	7	Cure
16	80	M	Pneumonia	<i>D. pneumoniae</i>	ND ^a	9	7	Cure
17	42	F	Pneumonia	<i>D. pneumoniae</i> ^b	—	6	8	Cure
20	39	M	Pneumonia	None	—	9	9	Failure
21	32	M	Pneumonia	<i>D. pneumoniae</i> ^b	—	6	5	Cure
						4	3	
22	47	F	Pneumonia	<i>Staphylococcus aureus</i> ^c	0.31	4	13	Cure
			Bacteremia	<i>Proteus mirabilis</i> ^d	10.0			
24	56	F	Pneumonia	<i>D. pneumoniae</i> ^b	—	6	6	Cure
25	59	F	Pneumonia with bacteremia	<i>D. pneumoniae</i>	0.04	9	17	Cure

^a Not done.^b Suspected pathogen.^c Isolated from sputum.^d Isolated from blood.

TABLE 3. Miscellaneous infections treated with cephacetrile

Patient no. ^a	Age (years)	Disease	Pathogen	MIC ($\mu\text{g/ml}$)	Daily dose(g)	Days of therapy	Clinical outcome
7	48	Bacteremia	<i>Staphylococcus aureus</i>	1.25	16 8	1 5	Patient ex- pired
15	21	Acute pyelo- nephritis	<i>Escherichia coli</i>	10.0	6 4	4 4 ^c	NF ^b
19	19	Acute pyelo- nephritis	<i>E. coli</i>	5.0	6	6 ^c	NF

^a All three were female.^b Patient did not return for follow-up.^c Received follow-up therapy with cephalixin.

the latter 12 patients who had discomfort with injections, 1, 3, and 8, respectively, had erythema, induration, and tenderness after 5 or more days of therapy. The complaints of pain were less frequent when the drug in solution was warmed to room temperature before administration and injected slowly. The local reactions subsided promptly when the site of injection was switched. One patient with mild pain requested intravenous administration after seven intramuscular injections.

Among the eight patients who received cephacetrile, intravenously, three developed mild phlebitis characterized by erythema, swelling, and tenderness at the site of the scalp vein needle. The phlebitis promptly resolved with a change in intravenous site. Several patients complained intermittently of pain with infusion of

cephacetrile, but after warming of the solution to room temperature before administration there were no further complaints.

Possible adverse reactions. Thrombocytosis was the most frequent laboratory abnormality observed during cephacetrile therapy. Nine patients developed platelet counts higher than normal. One patient was noted to have a platelet count lower than normal 1 month after cessation of therapy; 1 month later it had returned to normal. During therapy, eight patients developed modest increases in SGPT ranging from 40 to 100 units/100 ml (normal range: 0 to 20 units/100 ml for females and 0 to 40 units/100 ml for males). The SGPT returned to normal after treatment was stopped in four patients; three patients did not return for follow-up; and the enzyme was still elevated in the eighth patient when last seen 4

days after therapy was stopped. An increase in eosinophil count during therapy was noted in five patients. The direct Coombs' test became positive during the second week of therapy in four patients. Data are not available regarding reversion of the direct Coombs' test to negative in this group of patients. Leukopenia was observed in one patient (no. 6) during therapy and persisted during 4 months of follow-up; further evaluation of her hematological system, including bone marrow examination, was unrevealing. One patient (no. 25) developed a severe pneumonia due to an *Enterobacter* species 2 days after cephacetrile therapy was discontinued. This represented the only instance of superinfection in the 27 patients.

Drug susceptibility. The minimal inhibitory concentrations (MIC) of cephacetrile for the 25 initial pathogens isolated from 20 of the 27 patients are shown in Tables 1-3. The correlation coefficients relating the broth dilution values to zone sizes obtained with discs containing 60, 30, 25, 10, or 5 μg of cephacetrile were -0.807 , -0.864 , -0.894 , -0.883 , and -0.857 , respectively. Thus, the highest correlation was observed with discs containing 25 μg of cephacetrile. The regression line obtained with the 25- μg disc for the several groups of initial pathogens is shown in Fig. 2.

Serum concentrations. Cephacetrile concentrations were determined in the serum of 15

patients and are summarized in Fig. 3. A 1-g intramuscular dose in nine patients resulted in an average peak serum concentration of 22.7 $\mu\text{g}/\text{ml}$ 1 hr after administration which declined to an average of 1.65 $\mu\text{g}/\text{ml}$ 4 hr after injection.

After intravenous infusion of 1 g in 30 min, the average peak serum concentration in three patients was 69.4 $\mu\text{g}/\text{ml}$ at the end of infusion and was 0.52 $\mu\text{g}/\text{ml}$ at 4 hr. In three patients who received 1.5 g of cephacetrile intravenously, the average serum concentrations were 104.0 and 6.04 $\mu\text{g}/\text{ml}$ at the end of infusion and at 4 hr, respectively.

Urine concentrations. Cephacetrile concentrations in urine were determined in 15 patients during ongoing therapy. After a 1-g dose, the average urine concentration during the first 2 hr was 6.13 mg/ml, or a mean total urinary excretion of 99.3% of the administered dose; levels during the second 2 hr averaged 3.86 mg/ml, or a mean total urine output of 44.1% of the dose. After a 1.5-g dose, mean concentrations for the periods from 0 to 2 and 2 to 4 hr were 5.81 and 6.12 mg/ml, respectively; the mean excretion rates were 61.7 and 84.3% of the administered dose, respectively. The route of administration did not appear to effect the urine levels of drug.

DISCUSSION

When the activity of cephalothin, cephaloridine, and cephacetrile were compared against

FIG. 2. Regression line for cephacetrile 25- μg disc inhibition zones versus minimal inhibitory concentrations for 25 clinical isolates. Symbols: ●, *Staphylococcus aureus*; ○, *S. epidermidis*; ○, *Streptococcus* species; ■, *Enterobacteriaceae*; □, *Diplococcus pneumoniae*; □, *Haemophilus parainfluenzae*.

FIG. 3. Cephacetrile serum concentrations in 15 patients: means and ranges after intravenous or intramuscular administration. Symbols: ■, 1.5 g intravenously, 30-min infusion; ▲, 1.0 g intravenously, 30-min infusion; ●, 1.0 g intramuscularly.

50 clinical isolates of *S. aureus*, *S. faecalis*, and *Proteus* species, the mean MIC values were of the same order of magnitude for each antibiotic (6). However, against *E. coli* strains, cephacetrile and cephaloridine were 5 to 10 times more active than cephalothin. The susceptibilities to cephacetrile of the initial pathogens in the present study were similar to those previously reported (6). Thus, on the basis of the available in vitro data, cephacetrile would be expected to be efficacious in treating infections in which a parenteral cephalosporin was indicated. The clinical results in the present study establish early evidence of the effectiveness of cephacetrile. In particular, 12 of 16 patients with acute and extensive soft tissue infections were cured; 4 showed significant clinical improvement but their lesions resolved incompletely. Seven of eight patients with respiratory tract infections had clinical cures; one patient showed no response, but he did not have a bacterial infection. The two patients with urinary tract infections had prompt initial responses and probable cures, but follow-up was incomplete. The only patient who was designated as a therapeutic failure showed favorable clinical response with resolution of sepsis and negative blood cultures at the time she expired unexpectedly. The cause of her death is unknown, and a final assessment of the efficacy of cephacetrile could not be made.

Cephalothin and cephaloridine have been associated with pain on intramuscular injection and pain and phlebitis when given intravenously (1, 5, 7, 12, 17). Both of these complications were observed with cephacetrile administration. Although 11 of the 21 patients receiving cephacetrile intramuscularly had mild pain with injections and a twelfth had more severe pain, the complaints necessitated change to the intravenous route in only a single instance. Because complaints and local reactions were being solicited and examined for carefully in these patients, a final assessment comparing patient acceptance and tolerance of cephacetrile to cephalothin and cephaloridine cannot be made objectively.

The laboratory abnormalities of eosinophilia (7, 19), transaminase elevations (14, 16), and direct Coombs' test positivity (2, 3, 8, 11) have been observed with the two other parenterally administered cephalosporins. Thrombocytosis, observed in nine patients, may normally occur during convalescence following an acute infection and thus was not necessarily related to the cephacetrile therapy (9). Superinfection, which developed in one patient in this study, has been previously reported with cephalosporin therapy (1, 16).

The average peak serum concentration of

cephacetrile after a 1-g intramuscular injection in patients with acute infections in this study was lower than reported after a 1-g injection of cephaloridine in similar patients (1), but was higher than those observed after cephaloridine or cephalothin in healthy volunteers (13). The mean peak serum concentration of cephacetrile after intravenous administration was distinctly higher than those observed after comparable doses of cephalothin or cephaloridine in healthy volunteers (4). Similarly, the urine levels of cephacetrile were as high or higher than those seen with comparable doses of cephalothin or cephaloridine in healthy subjects (4). The total urinary excretion rates of cephacetrile for 4-hr periods in the present study possibly exceeded 100% of administered doses in part because of drug accumulation.

In summary, cephacetrile is an effective antibiotic which shares many of the properties of the presently available cephalosporins. Its place within the cephalosporin group will only be decided after further clinical information becomes available and its comparative efficacy and incidence of side effects can be assessed in a double-blind controlled study with other parenteral cephalosporins.

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