REFERENCES

1. Current, W.L., Reese, N.C., Ernst, J.V., Bailey, W.S., Heyman, M.B. and Weinstein, W.M. Human Cryptosporidiosis in immunocompetent and immunodeficient persons. N. Engl. J. Med., 1983; 308 : 1252.
2. Casemore, D.P., Sands, R.L. and Corry, A. Cryptosporidium species a "new" human pathogen. J. Clin. Pathol., 1985 ; 38 : 1336.
3. Tzipori, S. Cryptosporidiosis in animals and humans. Microcobiol. Rev., 1983; 47 : 86.
4. Cryptosporidiosis; assessment of chemotherapy of males with acquired immunodeficiency syndrome (AIDS). M.M.W.R., 1982; 31 : 589.
5. Cryptosporidium in the elderly; a cause of life-threatening diarrhoea. Am. J. Med., 1989; 86 : 507.
6. Zierdt, W.S. Concentration and identification of Cryptosporidium sp. by use of a parasite concentrator. J. Clin. Microbiol., 1984; 20 : 860.
7. Garcia, L.S., Bruckner, D.A., Brewer, T.C. and Shinizu, R.Y. Techniques for the recovery and identification of Cryptosporidium oocysts from stool specimens. J. Clin. Microbiol., 1983 ; 18 : 185.
8. Cryptosporidium spp. and Acromonas Spp. two new gasteroenteric pathogens in immunocompetent children. (Germ) Desgrand Champs D. and Munzinger, J. Hygienisch-Mikrobiologisches Institute. Kantonspp: tal. Ch-6000 Luzern 16 SWI-SCHWEIW MED. WOCHEWSCHR, 1989. 119/9 (276-281) Summ. In ENGL. FREN.
9. Paik, G. and Suggs, M.T. Reagents, stains and miscellaneous test procedure, in manual of clinical microbiology. 2nd ed. Washington, American Society for Microbiology, 1974, p. 930.
10. Fletcher, A., Sims, T.A. and Talbot, I.C. Cryptosporidial enteritis without general or selective immunodeficiency. Br. Med. J., 1982; 285 : 22.
11. Malla, N., Sehgal, R., Ganguly, N.K. and Mahajan, R.C. Cryptosporidiosis in children in Chandigarh. Indian J. Med. Res., 1987; 86 : 722.
12. Jokipli, L., Phjola, S. and Jokipli, A.M. Cryptosporidium; a frequent finding in patients with gasterointestinal symptoms. Lancet, 1983; 2 : 358.
13. Hayes, E.B., Matte, T.D., O' Brien, T.R., et al. Contamination of a conventionally treated filtered public water supply by cryptosporidium associated with a outbreak of cryptosporidiosis. N. Engl. J. Med. (in press).
14. Navin, T.R. and Juranek, D.D. Cryptosporidiosis; clinical, epidemiologic and parasitologic review. Rev. Infect. Dis., 1984; 6 : 313.

SHORT REPORTS

ANTIBACTERIAL ACTIVITY OF COPPER-AMINO ACID COMPLEXES

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ABSTRACT

Copper complexes of L-alanine, L-arginine, L-histidine, L-lysine, L-proline and L-threonine were studied for their antibacterial activity against Staphylococcus aureus, Streptococcus pyogenes and Escherichia Coli. The complexes of l-alanine, l-proline and l-threonine were nearly as active as ampicillin against Strep. Pyogenes. Mixed complexes of these amino acids showed similar effect. Other complexes were also active to a significant extent against all the three strains studied (JPMA 40 : 221, 1990).

Copper-amino acid complexes have been shown to possess anti- inflammatory (AI) activity[1,2]. The mode of action of such complexes is not yet clear. However, in the past some of the AI drugs used have been those basically developed as antibacterials. For example gold compounds used in the treatment of rheumatoid arthritis inhibited the growth of tubercule bacilli[3], Depsone[4], Indomethacine[5], Levamisol[6] and Pencillamine[7] being standard prescriptions to combat severe rheumatoid arthritis, are known to have antibacterial activity. Some of the copper complexes have also been studied against mycoplasma infections but no systematic study has been carried out in this regard[8,9]. In this communication we report a study of the antibacterial activity of copper-amino acid complexes to understand their mode of action.

MATERIAL AND METHODS

Copper-amino acid complexes were prepared in solution form according to the usual method[10,11] by mixing 2m mol of amino acid and 1m mol of copper acetate in water. The blue solutions thus obtained were used for

TABLE. Antibacterial activity of copper-amino acid complexes and their mixtures.

Compounds	Susceptibility Zone(mm)		
	E. Coli	Staph. aureus	Strep. pyogenes
Ampicillin	17	20	20
Copper Acetate	9	8	10
L-Alanine	R	R	R
L-Arginine	R	R	R
L-Histidine	R	R	R
L-Lysine	R	R	R
L-Proline	R	R	R
L-Threonine	R	R	R
$Cu(L\text{-}Alaninate)_2$	15	15	17
$Cu(L\text{-}Argininate)_2$	9	8	10
$Cu(L\text{-}Histidinate)_2$	9	8	10
$Cu(L\text{-}Lysinate)_2$	11	8	15
$Cu(L\text{-}Prolinate)_2$	15	16	18
$Cu(L\text{-}Threoninate)_2$	15	15	18
$Cu(L\text{-}Alaninate)_2 + (L\text{-}Prolinate)_2$	15	15	18
$Cu(-Alaninate)_2 + (-Threoninate)_2$	15	15	18
$Cu(L\text{-}Prolinate)_2 + (L\text{-}Threoninate)_2$	15	15	18
$Cu(L\text{-}Alaninate)_2 + (L\text{-}Prolinate)_2 + (L\text{-}Threoninate)_2$	15	15	18

R = Resistant

testing antibacterial activity. The mixtures of copper complexes were prepared by mixing two parts of amino acid and one part of copper salt in water. The amino acids used were L-alanine, L-arginine, L-histidine, L-proline and L- threonine.

The solution of the complexes were tested for their antibacterial activity both individually and as admixtures. The complexes of L-alanine, L-proline and L-threonine used in admixtures were selected because they showed significant activity when tested individually. In each case the solution containing equivalent to 10 µg of the complex was loaded on the susceptibility disc and the activity was measured against Staph. aureus, Strep. pyogenes and E. coli in the blood agar and Muller Hinton medium using ampicillin (10 µg) disc as standard according to the standard susceptibility testing method[12]. The sensitivity of copper acetate and the amino acids (10 µg) each was measured separately.

RESULTS AND DISCUSSION

The sensitivity results are listed in Table. All the amino acids tested were resistant. The activity shown by most of the copper-amino acid complexes is greater than that of copper salt indicating that the complexes possess significant antibacterial activity against the strains tested. The copper complexes of alanine, proline and threonine showed antibacterial activity only against streptococcus pyogenes comparable to that of ampicillin, whereas complexes of arginine, histidine, lysine showed less activities against these strains. The three mixed complexes of alinine, proline and threonine have also shown similar effects. From these results it appears that the hypothesis postulating the partial role of antibiotic activity of AI drugs in combating inflammation may be substantive. The knowledge of antibacterial activity of the copper-amino acid complexes gained through this study, and their possible effectiveness in inflammation strengthens the speculated role of bacterial pathogens in such type of ailment particularly in rheumatoid arthritis[13], and also provides an insight into their mode of action.

REFERENCES

1. Jackson, G.E., May, P. M. and William, D. H. Metal-Ligand complexes involved in rheumatoid arthritis-1. J. Inorg. Nucl. Chem., 1978; 40 : 1189.
2. Sorenson, J.R.J. Inflammatory diseases and copper. New Jersy, Humana Press, 1982, p. 289.
3. Koch, R. Antibacterial action of gold salts. Deuttsche. Med. Woreheusher, 1927; 16 : 756.
4. Toodman, L.S. and Gilman, A. The pharmacological basis of therapeutics. 4th ed. New York, Macmillan, 1970, p. 1311.
5. Plant, J.E., Higgs, G.A. and Easmon, C.S. Effect of antiinflammatory agents on chronic Salmonella typhimuriam infection in a mouse model. Infect. Immun., 1983; 42 : 71.
6. Lozada, F., Silverman, S. and Cram, D. Phemhigus vulgaris. A study of six cases treated with levamisole and prednisone. Oral. Surg., 1982; 54 : 161.
7. Vinnik, L. A., Vishnevetskii, F. E. and Perbukhin, Iu. V. Effect of D-Pencillamine on experimental tuberculosis. Probl. Tuberk., 1980; 3 : 64.
8. Brown, T.M., Baily, J.S., Iden, I.I. and Clark, H.W. Inflammatory diseases and copper. New Jersey, Humana Press, 1982, p. 391.
9. Mukhammud Zhanov Kh, R. Effect of some Trace Elements complexes on, E. Coli and Staph. aureus. Med. Zh. Uzb., 1985; 3 : 45.
10. Aberhalden, E. and Schnitzler, E. Copper salts of amino acids and polypeptides. Z. Physiol. Chem., 1927 ; 163 : 94.
11. Sorenson, J. R. Copper chelates as possible active forms of the antiarthritic agents. J. Med. Chem., 1976 ; 19 : 135.
12. Bauer, A. W., Kirby, W. M. M., Sheris, J.C. and Turck, M. Antibiotic susceptibility testing by standardized single disc method. Am. J. Clin. Pathol., 1966; 45 : 493.
13. Pullar, T., Hunter, J.A. and Capell, H. A. Which Component of sulphasalazine is active in rheumatoid arthritis? Br. Med. J., 1985; 290 :1535.

IFOSFAMIDE IN SOFT TISSUE SARCOMA

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Ifosfamide, a congener of Cyclophosphamide[1], was originally introduced into clinical practice in 1971, but has not been widely used because of severe dose limiting urotoxicity, particularly when used as a single agent in large doses. A resurgence of interest in Ifosfamide followed the demonstration that urotoxicity could be lessened or abrogated by fractionated administration[2] or, more effectively, by the concurrent administration of sulphahydryl compound mesna (2- mercaptoethanesulphonate), a highly uroprotective agent[3]. It is now clear that Ifosfamide has a broad spectrum of activity in human cancer[1-4]. This study reports our experience with Ifosfamide in patients with soft tissue sarcomas.

PATIENTS AND METHODS

All 14 patients (13 males 1 female) entered into the study had histologically diagnosed 'soft tissue sarcoma'. Nine of them were previously treated with chemotherapy and radiotherapy. Ages of the patients ranged from 2 to 65 years. Seven cases had rhabdomyosarcoma, 2 soft tissue sarcoma (NOS), 2- neurofibrosarcoma, and one each had fibrosarcoma, epithelimoid sarcoma and myxosarcoma.

Nine patients had extensive metastatic disease at the time of treatment. Lung metastases and inguinal lymphadenopathy were the most common manifestations.

Ifosfamide was given initially as a 5-day continuous infusion at a dose of 1.5gm/sqm daily. Mesna was given as