CLINICAL USE OF COMBINATIONS OF ANTIBIOTICS

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CLINICAL USE of combinations of ANTIBIOTICS

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PREFACE

Most physicians are in agreement that antimicrobial drugs are widely effective and fairly harmless. Most physicians also have the vague feeling that if one antimicrobial drug is good, two should be better, and three should cure almost everybody of almost every ailment. Emotions such as these probably contribute to the large scale abuse of combinations of antimicrobial drugs which must be held responsible, in part, for the rise of antimicrobial resistance among microorganisms, and for the high incidence of side effects to drugs among patients.

Nevertheless, it is necessary that physicians consider the use of drug combinations with its attendant advantages and disadvantages. There are clear-cut situations where the simultaneous use of two or more antimicrobial drugs is essential to the survival of a patient or the eradication of an infection. There are also unequivocal instances where a drug combination may be detrimental to a patient. The benefits and indications for the rational use of drug combinations change from time to time as new patterns of infection and of host response evolve.

It is fortunate, therefore, that Dr J. Klastersky was willing to organize a meeting for the systematic discussion of the pros and cons of antibiotic combinations. Dr Klastersky himself has contributed very importantly to this field by performing controlled clinical studies on the merits of combined chemotherapy in patients with severe impairment of anti-infectious defences. I am certain that the participants in this conference through presentations and discussions will contribute in an important way to a clarification of the problems of antibiotic combinations. I also hope that the deliberations of the conference may have a favourable impact on rational antimicrobial therapy through the world.

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I

COMBINATIONS OF ANTIBIOTICS – AN INTRODUCTORY REVIEW

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I have been asked to substitute for Dr Ernest Jawetz, who greatly regrets that he could not be present with you today. Dr Jawetz has contributed greatly to our knowledge of the effects of antibiotics in combination, and has been a perceptive critic of their use. I hope my presentation will do justice to him.

Antibiotics are too often prescribed for the psychologic satisfaction of all concerned. What other explanation can there be for the fact that 55 per cent of US physicians give antibiotics to patients whom they diagnose as having a common cold (46)? Combinations of drugs presumably have the virtue of giving more psychotherapy than single agents.

THE RATIONALE OF COMBINATION THERAPY

The main reasons for the use of antimicrobial agents in combination have changed little over past years, and have been well summarized (6, 16). They include (Table 1.1):

TABLE 1.1 The rationale of combination therapy

A	Initial therapy of serious infection
B	Treatment of infection due to multiple organisms
C	Decreased emergence of resistant microbes
D	Lessening the dose-related toxicity of treatment
E	Eradication of an infection which cannot be successfully treated with a single drug

A) Initial therapy of serious infection, to increase the likelihood that at least one effective drug is given at a time when either the organism or its antibiotic susceptibility is unknown. A single specific drug is selected as soon as better information is available. This situation arises most often in the newborn, in the post-abdominal surgery patient, and in patients with defective host defences (9, 40).

B) Treatment of infection due to multiple organisms, when it is unlikely that a single antimicrobial drug would be effective against all the species involved. However, control of these infections, e.g., peritonitis following a colonic perforation, usually depends more upon surgical drainage or debridement than upon antibiotics, although the latter may be important supplements.

C) Decreased emergence of resistant microbes where it has been shown that such resistance occurs frequently following the administration of single antibiotics. Resistant mutants are detected often if active pulmonary tuberculosis is treated with isoniazid or streptomycin alone (5). Resistant mutants were a serious problem when penicillin-resistant staphylococcal infections were treated with only erythromycin or novobiocin. The administration of gentamicin in conjunction with carbenicillin for infections due to *Pseudomonas aeruginosa* has been proposed specifically to decrease the selection of carbenicillin-resistant mutants (43).

D) Lessening the dose-related toxicity of treatment. The use of triple-sulphas avoids urinary blockage with crystals of sulphadiazine. A report that flucytosine and amphotericin B have at least additive effects *in vitro* and *in vivo* suggests a potential means of decreasing the prominent hazards of amphotericin treatment of systemic fungal infection (26, 49).

E) Eradication of an infection which cannot be successfully treated by a single drug, even though the drug is active *in vitro*. This justification of the use of antibiotic combinations usually involves the concept of 'synergism'. The therapist may seek to obtain killing or more rapid killing of organisms, rather than only an inhibitory effect. Alternatively, he may hope to increase a marginal antimicrobial effect to one with sufficient 'overkill' to sterilize the infected tissues. The best clinical documentation of the advantage of this type of antibacterial regimen is seen with enterococcal endocarditis (34). The same rationale is frequently offered in therapy of meningitis, osteomyelitis and other deep-seated serious infections, but our most pressing need for better clinical results is for the treatment of life-threatening infection in patients with impaired host defences, especially patients who are granulocytopenic.

DISADVANTAGES OF COMBINATION THERAPY

There are unfortunately potentially harmful effects resulting from the use of combinations of antimicrobial drugs (Table 1.2):

TABLE 1.2 Disadvantages of combination therapy

A	Suboptimal therapy for the actual pathogen
B	Excessive security leading to diagnostic delay
C	Difficulty of administration, leading to the use of plastic cannulae
D	Adverse drug reactions
E	Superinfection
F	Microbiologic antagonism
G	Chemical inactivation
H	Cost

A) Drugs selected for their broad spectrum will frequently be suboptimal for the specific organism in the particular case. We have seen pneumococcal meningitis develop in the face of cephalothin therapy for pneumonia, and others have reported the progression or development of meningitis during cephalosporin therapy (24).

B) Suboptimal therapy is particularly dangerous if the physician is deflected by the security of such 'coverage' – a favourite expression of my surgical colleagues – from pursuing a specific aetiologic diagnosis. Precious days and sometimes life are often lost, at least in part, because of delay while the attendants optimistically await the effects of whatever antibiotic 'cocktail' is in vogue.

C) A multiplicity of drugs, particularly those used in high dosage and associated with a significant incidence of phlebitis, may be physically difficult to administer. The patient may run out of superficial veins, and resultant use of indwelling plastic catheters invites bacteremic superinfection. Thirty-four cases – 18 per cent of the bacteremic infections acquired at our institution last year – arose from such intravascular devices.

D) Adverse drug reactions increase with the number of drugs administered. Over 13 per cent of all patients admitted to the Johns Hopkins Hospital in two months developed an adverse drug reaction, and antibiotics caused more than twice as many reactions as any other class of drugs except cardiotropic agents (42). The need for the sulphonamide component of co-trimoxazole has been questioned because many infections respond to trimethoprim or to sulphonamides alone, and the high incidence of toxic reactions in some reports appears to be due to the sulphonamide (2). When the febrile patient fails to defervesce or develops new fever after an apparent response, the diagnostic dilemma of uncontrolled infection versus drug fever is complicated by antimicrobial polypharmacy.

E) Patients in hospital acquire new, R-factor-containing enteric flora from their environment (8). A combination of antimicrobial agents may minimize the likelihood that the original infecting bacterium will become resistant, but probably increases the likelihood that the patient's gut will be repopulated with organisms resistant to both agents. In our hospital during 1972–3, infection directly contributed to the deaths of 35 patients whose host defences were impaired by blood dyscrasia, chemotherapy, steroids or radiotherapy. Five patients (14 per cent) died with fungal infection, and all had received extensive prior antibacterial therapy.

F) Microbiologic antagonism of the good effects of one antibiotic by another, although readily demonstrable *in vitro*, seems not to be an important clinical phenomenon, with the very significant exception of the treatment of common forms of bacterial meningitis. Tetracycline or chloramphenicol used in addition to penicillin or ampicillin results in more than twice the mortality rate found for control groups treated with a penicillin alone (21, 25). The marginal efficacy of cephalothin in the treatment of meningitis might be even more readily overcome by the same antibiotics. A poorer clinical result is also seen if a urinary tract infection is treated with antagonistic drugs to which the organism is sensitive rather than with synergistic combinations (22). The long-known antagonism of chloramphenicol for streptomycin has recently been extended by demonstration of *in vivo* antagonism of chloramphenicol for gentamicin (13, 38).

G) One antimicrobial agent may inactivate another *in vitro* and possibly *in vivo*, and we cannot assume chemical compatibility simply because a visible precipitate does not form. Thus, carbenicillin unequivocally decreases the half-life of gentamicin. However, the rate of this chemical inactivation is much less than the normal biological half-life (23). If solutions containing the two drugs are freshly made before use or the drugs are administered separately (33), and if the dose schedule for patients in renal failure is that proposed by O'Grady (31) rather than that of Kunin (20), no clinically significant problem is seen.

H) Cost of pharmaceuticals is a significant health care issue all over the world today. Our hospital spends $120000, nine per cent of the drug budget, on a single cephalosporin drug. At another hospital, 1·8 per cent of all patients receiving antibiotics were given carbenicillin, which, however, accounted for 18 per cent of the cost of all antibiotics (11).

A few years ago, at a national meeting of antimicrobial agents, a prominent expert in infectious diseases recommended that every patient with a diagnosis of sepsis receive full doses of carbenicillin, in addition to whatever antibiotics were already being empirically given, pending the returns from laboratory studies. If every such patient indeed received the then recommended 48 g per day at over $3/g for only two days, the cost in the US alone of treating 250000 bacteremic patients annually would increase by nearly $100 million.

The cost of combination antimicrobial treatment is obviously greater than that for single agents. Fixed combinations of antimicrobial drugs were found to be of unproven value (30) and this determination led to a ban by the Food and Drug Administration on their sale in the US. Prior to the ban, which was upheld by the courts in 1972, one major drug manufacturer derived one-eighth of its sales from one such mixture. It remains to be seen whether the unique properties of co-trimoxazole truly warranted a relaxation of that ban, or only reopened Pandora's Box.

SYNERGISM

Any discussion of treatment with combinations of antibiotics finds us before the semantic labyrinth known as synergism. *In vitro* antibiotic susceptibility testing is, after all, a convenient artefact. An extreme of potential irrelevancy might be the determination of the minimal inhibitory concentration of amphotericin B against *Histoplasma capsulatum* in the hyphal state as measured after several days at 22°C, which would then be related to the serum concentration of the drug. This contrasts with the clinical problem due to the yeast phase at 37°C in circumstances where the drug is given daily, with the whereabouts of poorly soluble amphotericin B within an hour of its once a day infusion practically unknown.

As we look for an *in vitro* effect of two or more antibiotics which is greater than can be attained by either drug alone, shall we examine the rate of microbial inhibition or killing, or shall we consider the concentrations of each drug needed to obtain the effect? Testing a streptomycin-resistant enterococcus from a patient with endocarditis, we found no viable organisms after 24 hours' incubation with either penicillin G 6 μg/ml or the combination of penicillin G 6 μg/ml plus gentamicin 5 μg/ml. The minimum lethal concentration at 24 hours was identical

whether penicillin G was used alone or in combination with the aminoglycoside, and thus by one measure of effect there was no synergism. However, serial subculture of the test system at intervals prior to 24 hours revealed a marked difference in the speed with which the enterococci were rendered nonviable, by four hours in the case of the combination but beyond eight hours for penicillin alone. By a different definition then, synergism was found. 'Killing' is itself arbitrarily defined – injured cells removed from a hostile environment have greatly different recovery rates depending upon the medium in which the subculture is made and the time allowed for recovery. Inhibitory and bactericidal effects vary greatly with inoculum size, incubation time and medium (15).

Our laboratory reports *in vitro* synergism as a minimum four-fold decrease in concentration of *each* of two drugs from the concentration of the individual agents needed to obtain a specified effect. Lesser decreases are considered simply additive or indifferent, and greater than four-fold increases in effective concentrations are considered evidence of antagonism.

Mechanisms of synergism seem to be of three principal types (Table 1.3):

TABLE 1.3 Mechanisms of synergism

A Blockade of the successive steps in a metabolic sequence
B Inhibition of inactivating enzymes
C Facilitated entry of one drug by another

A) Blockade of the successive steps in a metabolic sequence, exemplified by the combination of sulphonamides with the folate antagonists, pyrimethamine or trimethoprim. Inhibition of folate synthesis is more complete than that attained with either component of the mixture, and organisms only inhibited by the individual drugs may often be killed (12). The use of these compounds will be more extensively discussed by Dr O'Grady.

B) Inhibition by one drug of an enzyme capable of destroying a second drug. Recent interest has focused on the enhancement by methicillin, cloxacillin and cephalosporins of the activity of penicillin G and ampicillin against beta-lactamase-producing gram-negative bacilli. The beta-lactamase-resistant drugs are themselves ineffective against the bacilli, but their high affinity for the enzyme prevents inactivation of the enzyme-susceptible, antibacterially active drugs (48, 37). Exceedingly high concentrations of both drugs are required to demonstrate the benefits of penicillin-methicillin or ampicillin-cloxacillin interaction, thus far limiting clinical application of such combination therapy to infections of the urinary tract (36). Another drawback to this theoretically attractive approach is that synergism is most readily observed with low bacterial numbers (7). Dr Sabath reports on this subject in a later contribution.

Resistance to chloramphenicol and aminoglycosides among gram-negative bacteria is due mainly to enzymatic inactivation mediated by R-factors, which suggests additional fruitful areas for synergism by enzyme inhibition, if only the inhibitors were available.

C) Alteration of a microorganism by one drug which facilitates access of a

second drug to the site of inhibition was first suggested for the effects of the combination of penicillin and streptomycin upon *E. coli* (32). This physical change has proved to be the means by which penicillins enhance the activity of aminoglycosides upon enterococci (27, 28). If resistance to the aminoglycoside is due not to a permeability barrier but to a ribosomal mutation, which can be predicted by *in vitro* ineffectiveness of concentrations of aminoglycoside alone greater than 2000 μg/ml, synergism of penicillin G with that particular aminoglycoside does not occur (45, 50). Other drugs which interfere with cell wall synthesis, including bacitracin, cycloserine, vancomycin, beta-lactamase-resistant penicillins and cephalosporins, and EDTA, similarly display a synergistic action with aminoglycosides (27). This mechanism perhaps explains the synergistic action of bacitracin and other drugs upon staphylococci (14), of cephalothin and kanamycin upon methicillin-resistant staphylococci (3), and of reported ampicillin- and carbenicillin-gentamicin synergism against a variety of gram-negative enteric bacilli and *Pseudomonas aeruginosa* (17, 4, 43).

Facilitated entry may underlie the synergism of polymyxins and sulphonamides for strains of serratia and proteus (10, 47). However, polymyxin-sulphonamide synergy occurs in L-forms of *Proteus mirabilis* and *Staphylococcus aureus*, and thus greater penetration into the cell may not be the only cause of an increased activity (29). The potentiation by polymyxin B of the antifungal effect of flucytosine may have a similar explanation (41, 26).

PRESENT CLINICAL USE OF SYNERGISTIC COMBINATIONS OF ANTIBIOTICS

With the exception of synergism, the arguments I have given for the use of combinations of antibiotic drugs represent special situations in which there is general agreement that the benefits outweigh the disadvantages. The value of synergism leading to a bactericidal effect in the treatment of endocarditis is also well-established.

We do not have adequate data to judge whether synergistic or even bactericidal therapy is necessary for optimal results in acute bacterial meningitis or acute osteomyelitis. The clinical problem is not very pressing – the organisms which commonly cause these conditions are almost always sensitive to and treated with bactericidal therapy. In chronic osteomyelitis, where a necrotic sequestrum is usually present, it is difficult to believe that bactericidal therapy could make very much difference in the ultimately surgically determined result.

Antimicrobial therapy fails most frequently in the immunologically suppressed or neutropenic patient. Many recent reports claim that combinations of antibiotics, particularly carbenicillin and gentamicin, have substantially improved the therapy of serious gram-negative infections in neutropenic patients (35, 40). Studies in rats, mice and monkeys have not supported these uncontrolled clinical experiences, perhaps because of differences in the nature and intensity of the model infection and in the dose and time of instituting therapy (1, 44, 39).

It is particularly pleasing, therefore, to have on record the results of a randomized trial conducted at the Institut Jules Bordet in which patients receiving combinations of antibiotics which displayed synergism *in vitro* responded significantly better

(18 per cent failures) than patients in whom no synergism was found (47 per cent failures) (18). A subsequent controlled study from this Institute of carbenicillin and gentamicin therapy, alone and in combination, gave similar results (19).

We need more such studies to define groups of patients who will benefit from antibiotic combinations and to determine the optimal duration of such therapy. For example, important cost benefits and perhaps diminished risk of resistant mutants, toxicity and allergy might result if combined therapy need only be continued for 48–96 hours in most cases.

What does the future hold? The use of combinations of antibiotics in high-risk patients is now widespread. At our institution, the mortality of infection once it occurs in immunoincompetent patients is 9·5 per cent, more than double the 4·0 per cent for non-immunosuppressed patients. The mortality of infection in the subgroup of patients with haematologic malignancy is 13 per cent. However, all groups have an identical 18 per cent mortality in bacteremic infections. The excess mortality in these defective hosts includes 13 of 35 deaths (37 per cent) which were due to candida, aspergillus, pneumocystis, toxoplasma and mycobacteria, alone or in combination with each other or with more usual pathogens. Our therapy has become sufficiently effective that our patients die less from infection than from superinfection, and perhaps not from the first superinfection but from the second. Because of the continued selection and emergence of resistant bacteria, the need persists to develop new pharmacologic agents and better methods of their administration. Therapy for progressively more exotic infections can be improved. However, durable progress is likely only if we learn to destroy target cells, whether malignant or immunologically active, in a specific manner rather than with a howitzer, meanwhile developing greater skills in granulocytophoresis and transfusion, and homotransplantation of bone marrow.

CONCLUSION

The theoretical and practical justifications for the administration of antibiotic combinations are balanced by real disadvantages. Limitations of laboratory models and understanding prevent our assigning definite clinical meaning to *in vitro* synergism or antagonism except for those few situations in which a small number of careful clinical trials have confirmed their importance. Further controlled observations of the value of antibiotic combinations are necessary to delineate their optimal use. New components of these combinations will be necessary as more resistant bacteria emerge from our fertile environment. Ultimately progress will come from our ability to avoid, not treat, these infections.

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