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The Possible Clinical Value of Rifampicin and Trimethoprim in Combination

Summary: Although rifampicin is active against virtually all pathogenic bacteria, acquisition of resistance means that it cannot be used alone for treating infections. We have shown that, in combination with trimethoprim, this handicap can be overcome. Not only is the emergence of resistance prevented by the presence of trimethoprim, but antibacterial synergy is often observed. By applying certain logical guidelines, we have been able to suggest an appropriate combination of rifampicin and trimethoprim, which is now being tried in the treatment of various injections. There is no evidence that the emergence of resistant tubercle bacilli will be encouraged by the use of rifampicin in this way: on the contrary, this risk seems extremely remote.

Zusammenfassung: Der mögliche klinische Nutzen der Kombination von Rifampicin mit Trimethoprim. Obwohl Rifampicin gegen praktisch sämtliche pathogenen Bakterien wirkt, ergibt sich aus der Tatsache einer Resistenzentwicklung, daß es zur Behandlung von Infektionen nicht allein angewandt werden kann. Wir haben gezeigt, daß dieser Nachteil durch die Kombination mit Trimethoprim ausgeglichen werden kann. Nicht nur wird die Ausbildung einer Resistenz in Gegenwart von Trimethoprim, verhindert, sondern oft ist ein antibakterieller Synergismus festzustellen. Unter Einhaltung gewisser Richtlinien kamen wir zur Empfehlung einer geeigneten Kombination von Rifampicin und Trimethoprim, die zur Zeit bei der Behandlung verschiedener Infektionen erprobt wird. Es gibt keinen Hinweis, daß es durch diese Anwendungsart von Rifampicin vermehrt zum Auftreten resistenter Tuberkelbakterien kommt, dieses Risiko erscheint im Gegenteil äußerst gering.

Introduction

Rifampicin is inhibitory at therapeutically obtainable levels for almost all pathogenic bacteria. However, it is the experience of several workers that, when rifampicin is given alone for treating infections, highly resistant organisms emerge (1, 2, 3). The selection of these organisms is rapid, so that therapy often fails. A possible way to overcome this problem may be to combine rifampicin with another antibacterial agent. This method has been shown to be highly effective in the treatment of tuberculosis.

Choice of a Suitable Drug for Combination with Rifampicin

The drug chosen for combination must fulfil all or most of the following criteria:

1. For reasons which will be discussed, it must have an elimination half-life in man which is considerably longer than rifampicin (which has a half-life of 3.5 hours).
2. It should have a spectrum which is as broad as possible.
3. It should be lipophilic and well distributed in man, to ensure that, like rifampicin, it enters most body compartments.
4. Bacterial resistance to the drug chosen for combination should be rare, and there must be no cross-resistance with rifampicin.
5. It must be compatible with rifampicin and should be suitable for oral and parenteral administration, preferably twice a day.
6. Synergistic activity with rifampicin is desirable, but an additive effect may be suitable.

Consideration of these criteria with regard to available antibacterial agents eliminates all but trimethoprim. Even this is not ideal, as trimethoprim is poorly active against *Pseudomonas aeruginosa* and anaerobes, and some recent reports suggest that resistance of other organisms may be increasing (this is *not* our own experience, among commonly isolated bacteria (4)). However, on theoretical grounds trimethoprim seemed overall a suitable "back-up" compound, remembering that rifampicin is the "killer" drug in the combination. The finding of synergy between rifampicin and trimethoprim for certain bacteria was encouraging. In this respect it is noteworthy that both drugs interfere with different steps of the DNA synthetic process.

In Vitro Synergy

We have undertaken a large study to investigate quantitatively the interaction of rifampicin and trimethoprim against commonly isolated bacteria of medical importance. We were encouraged by an earlier study which used a small number of strains and a qualitative method (5). The authors remarked upon the "interesting" interaction of trimethoprim and rifampicin against gram-negative bacteria. It soon became apparent that, where synergy did occur, it was not on the same scale as that between trimethoprim and sulphamethoxazole; therefore, to be sure of accuracy we had to develop a discriminating technique for its detection. This was why we considered it mandatory to use drug concentration steps smaller than those employed in the conventional doubling dilution technique. We also defined synergy by rigorous criteria (6, 7). All our results, (including previously unpublished material) obtained using 363 unselected strains are summarized in Table 1. Synergy was observed in 41% of the strains tested. Antagonism was observed only with two species, *Staphylococcus aureus* and *Neisseria gonorrhoeae,* but only at extremely low concentrations of rifampicin (< 0.02 µg/ml) so it is unlikely that this would have any clinical relevance.

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Table 1: Combined in vitro activity of rifampicin and trimethoprim. Data from: 6, 7, 8 and unpublished experiments

Synergy often observed		Synergy not uncommon		Synergy rare or additive only	Antagonism*
Streptococcus pyogenes Streptococcus faecalis	44/44†	Enterobacter spp.	3/13	Escherichia coli	Staphylococcus aureus
		Klebsiella aerogenes	14/40	Salmonella typhimurium	Neisseria gonorrhoeae
Proteus mirabilis Proteus morganii Proteus rettgeri Proteus vulgaris	52/72	Pseudomonas aeruginosa	9/20	Providencia stuartii	
		Bacteroides fragilis	7/19		
		Haemophilus influenzae	3/14		
Serratia marcescens	15/15				

* Only when rifampicin < 0.02 µg/ml.
† Strains for which synergy observed/strains tested.

Grüneberg and *Emmerson* (9) tested selected strains of bacteria isolated from urinary tract infections, including some resistant to trimethoprim or rifampicin or both. They used a different methodology (doubling dilutions) and an alternative definition of synergy, so their results cannot be compared with ours; 16% of the strains investigated showed synergy. *Farrell* et al. (10) also tested a selected group of 61 strains, all of which were resistant to gentamicin, and found synergy in 43%.

As a result of the work described above, the following species must be added to those appearing in Table 1 for which synergy has been reported: *Staphylococcus epidermidis, Citrobacter freundii, Flavobacterium* spp., *Acinetobacter* spp. and *Alcaligenes* spp. *Grüneberg* and *Emmerson* (9) pointed out that the extent of synergy most commonly observed in these studies (where the sum of the fractional inhibitory concentration lies between 0.6 and 0.7 has been defined by a study group (11) as "slight or no potentiation". However, we wish to point out that under these conditions, inhibition is occurring with each drug present only at one third its individual minimal inhibitory concentration. This might be regarded by some workers as being of importance. However, we would emphasize that too much stress should not be laid upon the results of in vitro synergy experiments carried out by the technique described above. In our opinion, the absence of antagonism is of much greater significance. Another point that should be made is that we have never been able to demonstrate synergy by killing curves, turbidimetric measurements or disk tests.

One report of in vivo synergy has been made (12), but we are reluctant to impute much importance to this type of experiment carried out in the mouse, as the pharmacokinetics of these drugs are known to differ in man and in the mouse.

Suppression of Resistance Emerging

An in vitro finding of much greater significance than that of synergy was found by *Kerry* et al. (6). This was that the emergence of resistance to rifampicin was dramatically reduced in the presence of *sub-inhibitory* concentrations of trimethoprim, a finding that has since been confirmed by *Arioli* et al. (12). Clearly, when trimethoprim was present at inhibitory levels, no rifampicin resistance can emerge. Results of studies on the effect of the single drugs and of the combination on resistance emergence in gut flora organisms would be very helpful in establishing the validity of these in vitro findings to the in vivo situation.

Pharmacokinetic Considerations

The crucial findings described in the preceding paragraph dictated the design of pharmacokinetic experiments. The object of these was two-fold:

1. to determine whether rifampicin and trimethoprim interfere with the handling of one another by the body.
2. to see how much latitude in dosage schedules was possible while avoiding a "trimethoprim gap", which is that situation where rifampicin is present alone. Clearly this would allow rifampicin-resistant mutants to emerge.

Acocella and *Scotti* (13) used 600 mg rifampicin + 160 mg trimethoprim (a ratio of 3.75 : 1), given once daily, and *Hamilton-Miller* and *Brumfitt* (14) tested 300 mg rifampicin + 160 mg trimethoprim, (a ratio of 1.88 : 1), the mixture being taken 12-hourly. Healthy volunteers were used, and the drugs were given in capsule form. There was no evidence of any pharmacokinetic interference between the two drugs, except that the half-life of trimethoprim fell to 70–80% of its usual value (8–10 hours) after seven days' treatment with the combination. Both groups of workers considered this finding to be connected with the well-known inducing effect of rifampicin on liver enzymes; further experimental studies will be required to quantitate any changes in biliary excretion of the two drugs occurring during the course of a seven day regimen.

The feared "trimethoprim gap" did not occur on either dosage schedule, but it appeared that the once daily dosage resulted in sub-optimal levels of rifampicin during the second 12-hour period, while on the twice daily regimen the rifampicin dosage was also too small. A new plan is to use a different combination in a total daily dosage of 900 mg rifampicin + 240 mg trimethoprim. In this case, the dosage scheme will be one tablet (300 mg rifampicin and 80 mg trimethoprim) in the morning and two tablets (600 mg rifampicin and 160 mg trimethoprim) in the evening, for clinical trial work. The combination is to be given the name Rifaprim (Dow-Lepetit).

All the available evidence suggests that there will be no toxicity or side-effects using this type of schedule for a seven day period.

Lack of Risk of Emergence of Rifampicin-Resistant, Mycobacterium tuberculosis

The proposed use of rifampicin for non-tuberculous conditions has met with some opposition. Rifampicin is such a useful drug in the treatment of tuberculosis – having been responsible for the dramatic shortening in the total length required for treatment in this disease (15) – that many chest physicians have firmly set themselves against its use outside tuberculosis (e. g. 16), for fear that rifampicin-resistant *M. tuberculosis* strains could emerge. This question was put squarely in a leading article in Lancet (17). This editorial was deliberately provocative and encouraged the tuberculous lobby to state their case. Strangely, however, no response was forthcoming in the letters to the Lancet. A single letter appeared on the subject agreeing with the editorial (18) but there was no dissent. Any lingering fears which clinicians may still have must have been dispelled by a recent paper by *Acocella, Brumfitt* and *Hamilton-Miller* (19), where it was shown that the incidence of strains of *Myobacterium tuberculosis* showing primary resistance to rifampicin is no greater in countries where rifampicin is used freely and often alone for non-tuberculous disease than it is in countries where rifampicin use is strictly controlled.

Although there are good grounds for believing that rifampicin-resistant *M. tuberculosis* strains arise when rifampicin is improperly used for treating tuberculosis, but not when used properly, this is secondary resistance. Again, no dissension to this paper was expressed and no figures disproving the thesis have appeared since.

Thus, it must be concluded that the case has been made beyond reasonable doubt that the risk of selecting rifampicin-resistant strains of *M. tuberculosis* by using rifampicin for extra-tuberculous indications for seven days is very small. When Rifaprim becomes more generally available, however, it is important that a running check be made on the sensitivity of *M. tuberculosis* isolated before and after its introduction. It would also be a sensible precaution to avoid treating chest infections with Rifaprim until any possible risk has been assessed in large numbers of patients under careful surveillance.

In this context it is interesting to draw a comparison with the use of streptomycin in the early 1950's. Due to the lack of antimicrobial agents at that time streptomycin had to be used for both tuberculous and non-tuberculous indications. Yet despite a widespread belief to the contrary the facts are that such usage did not cause the emergence of streptomycin-resistant strains of *M. tuberculosis*. A further point is that the mutation rate to streptomycin resistance in the tubercle bacillus is a hundred times higher than that for rifampicin (20).

Another interesting question which has to be answered is whether emergence of resistance of non-tuberculous bacteria will occur. Again, until the combination has been used the answer will not be known. A point which needs to be stressed is that careful laboratory control must be exercised to ensure that organisms treated are sensitive both to rifampicin and to trimethoprim.

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Discussion

Reeves: Do you intend to give a loading dose of trimethoprim?

Brumfitt: Because a loading dose adds to the complexity of the treatment we do not intend to use it.

Stille: The objections of the clinician against an extended use of rifampicin are based not only on the risk of resistance against *Mycobacterian tuberculosis*, but also on the potential side effects of the drug. It can cause hepatotoxicity, and also lead to hemolysis and renal insufficiency.

Brumfitt: Toxicity to rifampicin on the dosage schedule which I gave will occur only very rarely. The „flu syndrome" (described with intermittant rifampicin dosage) is not a very serious disease. Hepatotoxicity is a very rare hazard with rifampicin. Thrombocytopenia is extremely rare. I do not think the risk of sideeffect should detract from the use of the combination, but, like all drugs, improper use is dangerous.

Bergan: What is the rationale for making the evening dose double that given in the morning? The interval is usually shorter between the night and morning doses than between the morning and night. I would think that reversing the dosing scheme would be better.

Brumfitt: At night the urine flow is reduced, so that the peak concentration in the body will occur during this period of time. Dosage three times a day may cause bad patient compliance.

Solberg: Do you think that the good cell penetration of trimethoprim and rifampicin is of any clinical importance in the treatment of common infections? I think that it could be of importance in the treatment of tuberculosis for example and maybe also salmonellosis, shigellosis, and brucellosis. But what about other infections?

Brumfitt: This combination will be indicated for serious but not less severe infections. Rifampicin has remarkable properties and is readily excreted into bile, but into urine only above a certain crucial level.

Solberg: We have used rifampicin for the treatment of patients with phagocytic disorders and impaired intracellular killing of phagocytised bacteria, and we think that rifampicin is of some importance in these conditions.

Reeves: One of the points that worries me about toxicity is the drug interaction that you might get due to induction of microsomal liver enzymes. If this combination were to be used on a large scale, for example in the treatment of UTI, you might get serious interactions such as women becoming pregnant when taking anticonceptive steroids.

Brumfitt: I agree that women taking the pill would have to be warned.

Forsgren: In addition to the side-effects of rifampicin discussed, I think interference with the immuno-response by inhibition of the action of lymphocytes should also be added. These effects occur at therapeutical concentrations as found in in vitro and in animal experiments.

Acar: I agree that rifampicin is a good drug which can be used in combination, not only for the treatment of tuberculosis. But I do not believe in a commercially marketed, fixed combination because there are other very good combinations with rifampicin. The best combination depends on the strain one is dealing with. Polymixin and rifampicin, for example, have given very good results in gram-negative bacteremia caused by very resistant organisms.

Brumfitt: This is again a problem of patient compliance. If people have to take two drugs, there is a risk that they take two of the wrong tablets or get the doses mixed up. This cannot occur with a fixed combination. If all physicians who treat serious infections had to work out the ratio of the components, the possibilities of getting it wrong might be greater than the disadvantage of having a fixed combination.