BIOAVAILABILITY AND EFFICACY OF RIFAMYCINS IN TB TREATMENT

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Summary: According to data of minimum inhibitory concentrations determined on the MTB test H37Rv strain, generated plasma peak concentrations levels and blood bacteriostatic activity application of Rifamycin sodium salt has an advantages over the use of oral rifampicin.

Key words: rifamycin sodium salt, tuberculosis, pharmacokinetics.

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Introduction

Tuberculosis occupies a leading position among infectious disease by the magnitude of infection reservoir [1]. The history of creation of anti-mycobacterium medications began in the 40–s of the last century, when fundamental chemotherapeutic agents were synthesized and subsequently introduced to treatment protocols. Due to the necessity to increase the efficacy of therapy in patients with pulmonary tuberculosis in the period of the epidemics, the improvement of treatment protocols for tuberculosis remains a current issue. The research directions in searching for more effective treatment modalities are linked to the synthesis of new anti-mycobacterium medications, the development of novel delivery methods and, ultimately, return to intravenous application of already known medications [2]. The current fundamental anti-tuberculosis agents are isoniazid, rifamycins, pyrazinamide, ethambutol and their respective combinations. Whereas isoniazid prevails over rifamycins in terms of bactericidal effect, the priority in sterilising action (the ability to kill Mycobacterium tuberculosis (MBT) populations with slow reproduction) is taken by rifamycin derivatives [3, 4, 5].

Rifamycins were discovered in 1957 and are classified as macrolactam antibiotics, known as ansamycins. According to contemporary nomenclature the producing organism of rifamycins belongs to Amycolatopsis (Nocardia) mediterranei. It is of interest to note, that in the process of vital activity in the medium containing corn and soy, Nocardia mediterranei is simultaneously forming rifamycins A, B, C, D and E, which all have different properties. However among the aforementioned rifamycin complex rifamycin B is known to possess the greatest stability. It has no marked activity against gram-negative bacteria, and only via consecutive chemical transformations its is possible to obtain the compounds with a wide activity spectrum: the aqueous solution of rifamycin B under conditions of aeration converts into rifamycin S (with an increase in its biological activity); subsequently, it is reduced with ascorbic acid with formation of rifamycin-SV, which possesses the greatest biological activity and is the least toxic among the aforementioned derivatives. Subsequent introduction of a charged group in the process of chemical synthesis leads to increased polarity of rifamycin-SV, which, in turn, greatly enhances its bactericidal activity against gram-negative bacteria [4, 6, 7, 8].

The cellular target of rifamycins is RNA-polymerase (RNA-P). The high efficacy of rifamycin derivatives is related to their ability to inhibit the process of transcription in the bacterial cell. Rifamycin binds to RNA-P in the ratio of 1:1, preventing the onset of the chain of polynucleotides within RNA, rather its further growth. Rifamycin does not bind to the RNA-polymerase of nuclei in mammal cells and does not influence their respective synthesis. Therefore, ansamycins block the activity of procaryotic RNA-P enzymes, which catalyse RNA
biosynthesis; however, at the same time they do not possess the ability to bind with eucariotic RNA-polymerase. All known mutations of resistance to rifamycins, found in RNA-P of various bacteria, are located in four areas of RNA-P β-subunit [9, 10, 11]. The most active rifamycin medications are its semi-synthetic derivatives, including Rifampicin and rifamycin SV sodium, which possess high activity against MBT and are used in the treatment of patients with pulmonary tuberculosis. The disadvantage of the latter is its low bioavailability in oral administration, which is compensated for by intravenous administration of the drug.

Rifamycins are mainly used in tuberculosis, although they are also highly effective in other inflammatory diseases, including those caused by multi-resistant staphylococci, acute and chronic osteomyelitis and infections of urinary and biliary systems. Rifamycins penetrate well into all tissues and body fluids, including cerebrospinal fluid. High concentrations are created in the sputum, pleural fluid and in the bones. Rifamycins are subject to biotransformation in the liver. In hepatic disease accumulation of these antibiotics may occur [12, 13, 14]. As of today, several formulations of rifamycin derivatives are registered in Ukraine (see Table 1).

<table>
<thead>
<tr>
<th>Trade name</th>
<th>Active substance</th>
<th>How supplied</th>
<th>Manufacturer</th>
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</thead>
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<tr>
<td>Makox</td>
<td>rifampicin</td>
<td>Capsules 150 mg each</td>
<td>Macleods Pharmaceuticals Limited, India</td>
</tr>
<tr>
<td>R-cin</td>
<td>rifampicin</td>
<td>Capsules 150 mg each</td>
<td>Lupin Limited, India</td>
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<td>Rifampicin</td>
<td>rifampicin</td>
<td>Capsules 150 mg each</td>
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<td>Lumière Pharma Ltd., Ukraine</td>
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<td></td>
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<td>JSC Pharmaceutical company Darnitsa, Ukraine</td>
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<td>Rifabutin</td>
<td>rifabutin</td>
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<tr>
<td>Rifapex</td>
<td>rifapentin</td>
<td>Capsules 150 mg each</td>
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<tr>
<th>Medications for parenteral application</th>
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<tr>
<td><strong>Rifonat®</strong></td>
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**Table 1. Formulations of rifamycin derivatives registered in Ukraine**

The studies of previous years have demonstrated that the antimicrobial effect of medications of the class under study depends exclusively upon their maximal concentration, whereas time of exposure to MBT does not play a decisive role. Taking that premise into consideration, we have performed a study of pharmacokinetics of medications in depending on administration route [15].

**Methods**
We have conducted a study of two formulations among ansamycins: rifamycin SV sodium, 30 mg/mL concentrate for preparation of solution for infusions and rifampycin, capsules for oral administration 150 mg each manufactured by Lupin Ltd., India.

During the study we have conducted:

- determination of serum concentrations of study medications by chromatographic method (using high-performance liquid chromatograph by "Perkin Elmer" Company, USA) during the study we have processed data from 80 blood samples collected in 20 patients;
- comparison of bacteriostatic activity of blood against laboratory H37Rv strain for 42 blood samples, obtained in 14 patients;
- survey of minimal inhibitory concentrations of oral rifampycin and rifamycin SV sodium medicinal agents on the Ploskauer-Beck liquid nutrient medium, complemented with normal horse serum ex tempore. The study was conducted upon H37Rv test strain.

The patients participating in the study received a single dose of rifamycin SV sodium of 450–600 mg by intravenous drip. In several days the patient received comparator medication, rifampycin in capsules in a single oral dose of 600 mg.

The choice of evaluation terms was based upon the data of pharmacodynamics of the medications and the peculiarities of MBT reproduction, as well as the necessity to create high peak concentrations of medications in blood, and, therefore, the relevant bacteriostatic activity of blood (BAB) for the successful treatment of patients with tuberculosis.

Two schedules of administration of medications were compared:

1. Rifamycin SV sodium intravenously in the Main Group (MG);
2. Rifampycin orally in Control Group (CG).

**Results**

Data of two patients among the studied are given on Figures 1 and 2.

**Fig. 1. Serum pharmacokinetics of medications in Patient A.**
Fig. 2. Serum pharmacokinetics of medications in Patient B.

In course of the study it was estimated that the maximal concentration of rifamycin SV sodium after intravenous administration is $22.9 \pm 2.3$ mcg/mL, which substantially exceeds the concentration of rifampicin in capsules when administered at the dose of $450 - 600$ mg ($8.9 \pm 1.3$) mcg/mL ($p < 0.05$). The data are presented at Figure 3.

Fig. 3. The peak concentrations of medications in various routes of administration
Note: *– the difference between the indices is significant ($p < 0.05$)

In course of the study it was estimated, that the minimal inhibitory concentration of Rifonat is 10 times smaller than the respective concentration of Rifampicin ($0.03$ mcg/mL and $0.3$ mcg/mL, respectively), the data are presented in Table. 2.
Medication | The concentration of study medication, mcg/mL
---|---
| 5 | 2.5 | 1.25 | 0.6 | 0.3 | 0.15 | 0.07 | 0.03 | 0.015 | Control
Rifampicin | – | – | – | – | – | + | +++ | ++++ | ++++ | ++++
Rifamycin SV sodium | – | – | – | – | – | – | – | – | ++ | ++++

Table 2. The minimal inhibitory concentrations against H37Rv strain, mcg/mL.

Notes:
– growth of MBT film absent
+ growth of MBT film present

During the first hour of study, intermediate and high BAB indices were found in 100% of cases in groups compared. In MG high 1st hour BAB indices were significantly more frequent, in 87.5 ± 11.7% patients, compared with CG, where the index acquired comparable values in 33.3 ± 19.2% patients (p < 0.05).

Repeated detection of BAB levels was performed in 3 hours after administration of medications. In 3 hours of observation high BAB levels were found in 62.5 ± 17.1% MG patients and intermediate levels were present in 37.5 ± 17.13% in patients. In application of oral rifampicin high BAB levels were detected in 50.0 ± 20.4% of CG patients. It should be noted that although overall intermediate and high levels of BAB were noted in 100% cases in both MG and CG groups, the proportion of patients with high levels of BAB was greater in MG, where intravenous administration of rifamycin SV sodium was applied. Data presented in Table 3.

<table>
<thead>
<tr>
<th>Hour of study</th>
<th>Groups of patients</th>
<th>The values of bacteriostatic activity of blood</th>
</tr>
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<tr>
<td></td>
<td></td>
<td>Low 1:2 – 1:4</td>
</tr>
<tr>
<td></td>
<td>abs</td>
<td>%</td>
</tr>
<tr>
<td>1</td>
<td>MG(8)</td>
<td>–</td>
</tr>
<tr>
<td></td>
<td>CG(6)</td>
<td>–</td>
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<tr>
<td>3</td>
<td>MG(8)</td>
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<td></td>
<td>CG(6)</td>
<td>–</td>
</tr>
<tr>
<td>6</td>
<td>MG(8)</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td>CG(6)</td>
<td>2</td>
</tr>
</tbody>
</table>

Table 3. Bacteriostatic activity of blood against H37Rv strain, (M±m%)

Note * – The difference between the indices of 1st and 2nd groups is significant (p < 0.05).

At the 6th hour of study the data of MG and CG groups were characterized by further decrease of bacteriostatic activity of blood and its detection at low levels in 37.5 ± 17.1% and 33.3 ± 19.2% patients, respectively. The patients in oral rifampycin group did not have high levels of BAB in any case; however, intermediate levels of the index were present in 66.7 ± 19.2%
patients. Intravenous administration of rifamycin SV sodium by the overall values of intermediate and high BAB indices was somewhat lower compared to the group where rifampicin was applied orally 62.5 ± 17.1% and 66.7 ± 19.2%, respectively. However, in this group in 50.0 ± 17.7% cases high BAB levels were detected, unlike CG, where high BAB were not registered at all.

Bacteria excretion in patients of both groups is shown on the figure 4.

**Fig.4** Bacteria excretion in the patients of the main (rifamycin SV sodium) and control (rifampicin) groups.

The average time of abacillation in sputum in the main and control groups in patients with advanced pulmonary tuberculosis were (1.6 ± 0.1) and (2.1 ± 0.2) months, respectively (p <0.05).

The average healing time of healing of cavities in the main group was (3.2 ± 0.2) months vs. (3.9 ± 0.3) months in the control (p> 0.05).

**Discussion and conclusion**

The use of oral therapy in some cases cannot take into consideration several factors that decrease the dose of administered drugs, as well as their bioavailability (because of vomiting, diarrhea, the primary effect of passing through the liver, functional and morphological status of the gastrointestinal tract, etc.). Intravenous administration can achieve 100% bioavailability of drugs by eliminating the physiological factors that lead to its decrease. In intravenous infusion you can create the necessary concentration of anti-TB drugs depending on the rate of administration of drugs.

Since one of the ways to increase treatment effectiveness in patients with tuberculosis is intravenous administration of medications, in course of our further work we conducted comparison of different modes of administration of medications. Taking into account the data of study conducted, rifamycin SV sodium can be recommended as a medication of choice in treatment of patients with pulmonary tuberculosis (10 times more active against H37Rv strain compared to rifampicin), and its intravenous administration allows for creating high levels of bacteriostatic activity of blood and peak serum concentrations 2.5 times more than those in patients on oral rifampicin therapy. As intravenous chemotherapy with rifamycin SV sodium in the etiotropic treatment is superior to standard oral chemotherapy on the evidence of the data of pharmacokinetics, microbiological research and clinical observations, further research are
needed to evaluate the possibility to increase the effectiveness of treatment of TB patients with intravenous isoniazid, ethambutol and PAS – the most active and frequently used anti TB drugs.

References:


