

### Quinine is bactericidal

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#### Abstract

**Objective:** To determine the antibacterial properties of Quinine dihydrochloride and its MIC.

**Material and Methods:** A Quasi-Experimental study was conducted at the Jinnah Postgraduate Medical Centre, Karachi from July 2006 to November 2006. Two hundred samples of pus, blood, sputum and ascitic fluid, from hospitalized adult patients from Medicine ward (W-7) having different Bacterial infections were studied. Proforma was filled to document the demographic details. Samples were collected from different sites, isolated, identified and checked for antimicrobial susceptibility of quinine dihydrochloride by standard methods.

**Results:** Staphylococcus aureus, Escherichia coli, Proteus vulgaris, Pseudomonas aeruginosa and Salmonella typhi were inhibited at MIC of 125 g/ml. Streptococcus pyogenes was inhibited at MIC of 31.25 g/ml of quinine dihydrochloride (Dilution ranges from 31.25 g/ml to 1000 g/ml).

**Conclusion:** The concomitant antimalarial and antibacterial action of Quinine dihydrochloride may be beneficial in developing countries adding to cost effectiveness of treatment provided to patients belonging to low socio-economic group (JPMA 59:208; 2009).

#### Introduction

Quinine is a known antimalarial drug. It is the chief alkaloid of Cinchona, the bark of South American Cinchona tree. It is believed to have gotten its name from countess of Cinchon, wife of the Spanish Viceroy of Peru, who in 1638 fell desperately ill with malaria. Fortunately, she was cured by using the ancient herbal remedy of "quinquina bark" and in her honour, the tree was named Cinchona.<sup>1</sup>

Quinine is used for malaria since centuries, it acts principally on the mature trophozoite stage of parasite development, does not prevent sequestration and does not kill the pre-erthrocyte sexual stage of plasmodium falciparum.<sup>2</sup>

Initial therapy of severe malaria should always be parenteral. The recommended loading dose of quinine is 20 mg/Kg dissolved in 10 ml/kg of dextrose water or normal saline over a period of 4 hours.<sup>3</sup> Intramuscular quinine is another alternate or initial therapy, if facilities for, controlled intravenous quinine administration are not available.<sup>4</sup> The patient should be switched to oral quinine as soon as possible. If parenteral quinine has to be continued beyond 48 hours, or if renal failure supervenes, the maintenance dose should be reduced to 5-7 mg/kg to avoid quinine toxicity.<sup>2</sup> The oral dose is 10 mg/kg dose (maximum 600 mg) three times a day. Total duration of therapy is 7 days. The only contraindication of quinine is allergy and G6PD deficiency.<sup>5</sup> Jaundice, renal failure, hypotension and thrombocytopenia are not contraindications for quinine administration.<sup>6</sup>

Its medical application is throughout the world from

South America to Asia and Europe. Some of the conditions for which quinine has been utilized include, adenopathy, alcoholism, amoebiasis, anaemia, anaesthetic, appetite stimulant, arrhythmias, carditis, dandruff, diarrhoea, weakness, neuralgia, sorethroat, insecticide, leg cramps, haemorrhoids neuritis, antiviral antibacterial, typhoid and malaria.<sup>7</sup>

Antimalarials are primarily developed to treat malaria and quinine is well known for saving millions of patients with this disorder. Malaria is still a major cause of death in the tropics.<sup>8,9</sup>

Food Drug Administration (FDA) has approved antimalarials for malaria, systemic lupus erythematosus and rheumatoid arthritis. But interestingly antimalarials have recently been found to be effective as additional therapy for acquired immunodeficiency syndrome (AIDS).<sup>10</sup> Recently it has been discovered that quinine sulphate is also effective against Herpes simplex virus 1 (HSV1).<sup>11</sup>

Some of the patients who receive antimalarials for the treatment of systemic lupus erythematosus, rheumatoid arthritis and severe falciparum malaria are also immunosuppressed because of their disease and have concomitant bacterial infections. These patients often need systemic antibiotics either prophylactically or therapeutically for treatment of the infection. The antibacterial action of antimalarial drugs protection against or even treatment for the infection, it obviates the need for additional antibiotics.<sup>12</sup>

The purpose of our study was to determine the antibacterial effect of quinine dihydrochloride on gram

positive and gram negative bacteria in isolates obtained from the indigenous population.

### Material and Method

The study was conducted in the Department of Microbiology Basic Medical Sciences Institute (BMSI).

Two hundred samples from hospitalized adults patients having different bacterial infections like typhoid fever, diabetic foot, peritonitis and respiratory infection were randomly collected without discrimination of age, sex and race. Twenty-five isolates of each organism were used. The drug base was supplied by Pharmedic pharmaceutical company. The MIC of Quinine dihydrochloride were determined against the Gram positive bacteria, *Staphylococcus aureus* and *Streptococcus pyogenes* and Gram negative *Pseudomonas aeruginosa*, *E.coli*, *Proteus mirabilis* and *Salmonella typhi*. The MIC was performed by Broth Macro dilution method.<sup>13</sup>

The cultures of these organisms were collected from clinical specimens from Medical ward-7 and stock were taken and identified by standard methods. The media used were Blood agar (Merck KGaA 64271 Darmstadt, Germany), MacConkey agar (Merck KGaA 64271 Darmstadt, Germany), and Mueller Hinton broth (Oxoid Ltd, Basing Stoke, Hampshire, England). The stock solution was prepared in distilled water. The drug concentration in stock solution was 20000 g/ml. The working dilution ranges were prepared from the stock solution in Muller Hinton broth by double dilution method. The ranges of working dilution were 1000 g/ml, 500 g/ml, 250 g/ml, 125 g/ml, 62.5 g/ml and 31.25 g/ml. These were intermediated dilution of Quinine dihydrochloride concentrations.

The inoculum was prepared by adding 4 isolated colonies in the 4 ml Muller Hinton broth.<sup>14</sup> The prepared inoculum was compared with a 0.5 McFarland Standard in the Muller Hinton Broth (Annex-i) The inoculum was diluted 1:100 in Muller Hinton broth. The tests were performed in the 75X12mm sterile capped tubes. One ml of each drug dilution was transferred in the marked tubes. Then 1ml of test organism was added in each tube. The final concentrations formed were 500 g/ml, 250 g/ml, 125 g/ml, 62.5 g/ml, 31.25 g/ml, 15.625 g/ml (Anex-ii). The tubes were incubated at 37°C for 24 hours. After incubation period results were recorded on the basis of turbidity (Annex-ii).<sup>15</sup> This experiment on the clinical isolates was repeated three times with controls on the same isolates. Twenty-five of each strain types were used with 05 control organisms.

The inclusion criteria were: hospitalized patients who presented with clinical features suggestive of bacterial infections such as typhoid fever, diabetic foot, peritonitis, respiratory tract infections and wound

infections other than diabetic foot. Sample collected were pus, ascitic fluid, blood, and sputum. Patients were followed from their time of presentation in the out patient department till the time of their admission to the ward. Samples were collected prior to the commencement of treatment with antibiotics.

Known or suspected cases of pulmonary tuberculosis, known or suspected cases of malignancy particularly in patients with ascites and patients receiving any antibiotic treatment at the time of presentation were excluded.

### Results

A total of 200 clinically suspected cases of different diseases i.e. typhoid fever (14%), diabetic foot (26.5%), superficial skin infection (16.5%), severe respiratory tract infection (30%) and peritonitis (13%), attending out patients or admitted patients in Medical Ward-7, JPMC, Karachi, during the period July 2006 to November 2006 were included in this study.

Table 1 shows age distribution of study subjects. It was observed that most of the patients belonged to age group 31-40 years (32.5%), followed by 41-50 years (22.5%).

#### Annexure i

##### Preparation of McFarland Standard

1. One percent (1%) v/v solution of sulphuric acid was prepared by adding 1 ml of concentrated sulphuric acid to 99 ml of distilled water and mixed well.  
Caution: Concentrated sulphuric acid is hygroscopic and highly corrosive. Therefore, mouth pipetting was avoided and water is never added to the acid.
2. One percent (1%) w/v solution of barium chloride was prepared by dissolving 0.5 g of dehydrated barium chloride ( $\text{BaCl}_2 \cdot 2\text{H}_2\text{O}$ ) in 50 ml of distilled water.
3. 0.6 ml of the barium chloride solution was added to 99.4 ml of the sulphuric acid solution and mixed.
4. A small volume of the turbid solution was added to a capped tube or screw-cap bottle of the same type as used for preparing the test and control inocula.

The samples received were sputum (30%), pus from diabetic foot (26.5%), pus from superficial skin infection (16.5%), blood (14%), and ascetic fluid (13%) respectively.

Table 2 shows the minimum inhibitory concentration of quinine dihydrochloride against the organisms isolated.

### Discussion

Quinine is regarded on an antiviral, bactericidal and antiprotozoal in the old and recent literature.

The present study was conducted to see the

**Annexure ii**

<b>Stock solution</b>	<b>Working dilution</b>	<b>Intermediate concentration</b>	<b>Final conc:</b>	<b>MIC</b>
1ml+ 20000 g/ml	1ml+19ml	1000 g/ml	500 g/ml	S.aureus-NBG Strept:pyogenes.-NBG Psued:-aeruginosa-NBG E.coli-NBG Proteus-mirabilis -NBG Salmonella typhi NBG
2	10ml+10ml	500 g/ml	250 g/ml	S.aureus-NBG Strept:pyogenes.-NBG Psued:-aeruginosa-NBG E.coli-NBG Proteus-mirabilis -NBG Salmonella typhi NBG
3	10ml+10ml	250 g/ml	125 g/ml	S.aureus-NBG Strept:pyogenes.-NBG Psued:-aeruginosa-NBG E.coli-NBG Proteus-mirabilis -NBG Salmonella typhi NBG
4	10ml+10ml	125 g/ml	62.5 g/ml	S.aureus-NBG Strept:pyogenes.-NBG Psued:-aeruginosa-NBG E.coli-NBG Proteus-mirabilis -NBG Salmonella typhi NBG
5	10ml+10ml	62.5 g/ml	31.25 g/ml	S.aureus-NBG Strept:pyogenes.-NBG Psued:-aeruginosa-NBG E.coli-NBG Proteus-mirabilis -NBG Salmonella typhi NBG
6	10ml+10ml	31.25 g/ml	15.625 g/ml	S.aureus-NBG Strept:pyogenes.-NBG Psued:-aeruginosa-NBG E.coli-NBG Proteus-mirabilis -NBG Salmonella typhi NBG

BG=Bacterial growth\*

NBG= No bacterial growth

\* It was on the basis of turbidity

MIC= Minimum inhibitory concentration

antibacterial effect of quinine dihydrochloride against different gram positive and gram negative pathogenic organisms. It was observed that quinine dihydrochloride had bactericidal effects on Staphylococcus aureus, Streptococcus pyogenes, Escherichia coli, Proteus vulgaris, Salmonella typhi and Pseudomonas aeruginosa.

Our results are supported by those of Rennie et al.<sup>16</sup> who discovered that quinine sulphate inhibited Staphylococcus aureus, Enterobacter agglomerans, Klebsiella pneumonia and Pseudomonas aeruginosa.

Wolf et al.<sup>17</sup> found that quinine sulphate inhibited the

internalization/invasion of Escherichia coli.

In the presented study quinine dihydrochloride was found to be bactericidal but minimum inhibitory concentration (MIC) was high. It is presumed that quinine dihydrochloride may be more effective against pathogenic organisms in vivo at same MIC level required for malaria. Quinine dihydrochloride may obviate the need for additional antibiotic or antibacterial drugs.

The advantage of antimalarials is that, they not act directly on the invading pathogens, but rather on host cells, so that there are minimal, chances for microorganisms to

become resistant to it.<sup>12</sup>

## Conclusion

This study subsequently supports the antibacterial effects of Quinine dihydrochloride.

**Table 1: Age distribution.**

Age in years	N	Percent
15-20	15	7.5
21-30	40	20
31-40	65	32.5
41-50	45	22.5
51-60	25	12.5
61-70	10	5

The concomitant antimalarial and antibacterial actions of Quinine hydrochloride may be beneficial in a developing country for economic reasons where a dual effect is achieved by a single drug thus obviating the need of an additional antibiotic.

**Table 2: MIC of Quinine dihydrochloride against organisms.**

Name of organism	MICs values
Staphylococcus aureus	125 g/ml
Pseudomonas aeruginosa	125 g/ml
Escherichia coli	125 g/ml
Proteus mirabilis	125 g/ml
Salmonella typhi	125 g/ml
Streptococcus pyogenes	31.25 g/ml

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