Abstract: Bacterial resistance to antibiotic treatment is a very problematic issue, since introduction of any new antibiotic is shortly followed by the occurrence of resistant bacterial isolates in the clinic. The apparent increase in clinical resistance to antibiotics is particularly a distressed situation in nosocomial infections, where already defenseless patients can be unsuccessful to respond to treatment, causing even greater health issue. Nosocomial infections can be identified as those happening within 2 days of hospital acceptance, 3 days of discharge or 1 month of an operation. They influence 1 in 10 patients admitted to hospital. Annually, these outcomes in 5000 deaths with a cost to the National Health Service of a billion pounds. Although these problems, antibiotic therapy is still the most common procedure used to combat bacterial infections. Conversely, it is often said that the host’s immune system plays a primary role in the development of infections. Accordingly, we proposed a mathematical model defining population dynamics of the specific immune cells produced according to the properties of these by host and the bacteria exposed to multiple antibiotics synchronically, presuming that resistance is gained through mutations due to exposure to antibiotic. Qualitative analysis reveals of that equilibrium points giving important ideas about the proliferation bacteria and immune cells. According to the results of this analysis, there is that possible scenarios existing of bacteria, resistant bacteria and immune system cells and only resistant bacteria, when the antibiotic treatment is not enough to destroy the infection. These scenarios have expressed the persistent infection situation that immune cells do not respond to this infection or continue to exist with bacteria causing this infection in the host’s balance. On the contrary, if the antibiotic treatment is enough to destroy the infection, then there is the situation that the infection is removed. Furthermore, these results was supported by numerical simulations.

Keywords: Ordinary differential equations systems, Equilibrium points, Immune system response, Bacterial resistance, Multiple antibiotic therapy.

AMS Subject Classification 34K20, 92C50, 92D2

1. Introduction

It is expressed that infections have been the principal cause of diseases throughout the history of mankind [1]. There are particularly bacterial infections among these. The most prevalent method known to struggle against bacterial infection is through antibiotic therapy applied to individuals. The expression of resistance to antimicrobial agents in the therapy is both the logical and inescapable consequence of using these agents to treat human infections [2,3]. Resistance developed by the bacteria against antibiotics is identified as the ability of bacteria to resist the effects of antibiotics designed to eradicate and control them [4]. Shortly after the release of each new class of antibiotics for treatment, this has been followed by the emergence of new strains of bacteria which are resistant to this class [5-7]. In this context, the development of novel treatments for bacterial infections is of utmost importance [8].

It is expressed that antibiotics has the bacteriostatic action to stop the growth of bacteria and bactericidal action to wipe out the bacteria. But, this difference is not clear, because it depends on the drug concentration used and the growth stage and the species of bacteria [9]. Accordingly, the multiple antibiotic therapy is a more appropriate form of treatment.

Indeed, the bacterial infection is a complex continuum for both the infectious bacteria and the host [10]. It is proposed that the immune system plays fundamentally a significant role in the progress of infections [11].

The immune system is expressed as a system of biological structures and processes in an organism which protects the body from possible harmful substances by recognizing and responding to antigens [12]. For this reason, the reactions of different hosts against the same infection can be different due to immune system response given by host.

In light of the above, dynamics of relations among antibiotics, immune cells and bacteria are important to understand the nature of the infection. Mathematical models are one of the significant tools used in both analyzing the spread of infectious
diseases in a population of individuals [13,14], and predicting the timing and expansion of infection and possible reinfection processes in an individual [15]. While the former is usually used for planning, prevention and control scenarios, the latter can be active in therapy/intervention programs for treating the individuals exposed to the particular pathogen. In this respect, understanding and anticipating the time of occurrence and magnitude of the maximum load of the bacteria and immune system cells by mathematical modelling can be crucial in selecting effective intervention strategies [16]. Therefore, results on reproduction of sensitive and resistant bacteria to antibiotics are obtained in [17-21]; definitions of factors responsible for resistance prevalence are studied in [11,22-24]; bacteria behavior under different antibiotic treatments is examined in [25-30]; optimization results and design of control measures are investigated in [18,31-34]; biological cost and persistence of antibiotic resistance are analyzed in [1,35-38]; dynamics between pathogen and immune response are examined in [15,16,39-45], respectively. In this study, the effects on bacteria causing infection of both the multiple antibiotics therapy having Holling's response (2nd type) as well the immune system response was investigated through mathematical modeling. Therefore, it has tried to bring a different perspective from the studies made above. We have constructed a continuous time model considering the host's immune response and the basic mechanisms of bacterial resistance to antibiotics. Our aim is to obtain certain conditions dependent on the development of sensitive and resistant bacteria population under the pressure of immune cells and multiple antibiotics.

2. Model formulation

When the emergence of resistant bacteria to antibiotic has modelled, there are the two basic aspects generally considering. These are within-host models and within-hospital compartmental models. Within-host compartmental models are generally SIR models in epidemiology, considering especially how infections will expand throughout the hospital [9-11]. These models are useful to develop strategies to prevent the spread of resistant individuals to antibiotic in hospital. Current mathematical models focusing on the fact that the resistant development is in the host, aim to study how antibiotic treatment strategies can affect, and in addition, how the occurrence of antibiotic resistance can be prevented [12,13]. In such models, the effect of immune cells generated by the host under the pressure of the bacteria are frequently either neglected or assumed to be at a constant rate. We built up the mathematical model including cell-mediated immune response. In addition, treatment regimens involving multiple antibiotic have been used in most bacterial infections due to bacterial resistant. In this sense, besides the interaction of bacterial-immune response, to investigate the effects of multiple antibiotic therapy is biological more meaningful.

In our study, it has been suggested that the model where the arise of resistant bacteria to antibiotic is in host and so the load of sensitive and resistant bacteria to antibiotic, the effects of multiple antibiotic therapy and the immune system's response of host has examined. Our aim is to find specific parameters determining the change in the concentrations of the immune system's cells produced in host to fight these and the sensitive sub-populations and resistant sub-populations that has either arisen through random mutation and clonal enlargement or through cross-contamination in a special infection and under a appropriate treatment regimen.

We have analyzed the concentrations of the immune cells and bacteria in an individual receiving a cocktail of multi-drug treatment against bacteria via mathematical modelling. We have assumed that the population sizes of sensitive and resistant bacteria against multiple antibiotics at time t denote by $S(t)$ and $R(t)$, respectively. Moreover, let us identify by $B(t)$ population sizes of immune cells at time $t$ and by $A_i(t)$ the concentration of the $i$-th antibiotic for $i = 1,2,\ldots,n$ at time $t$. The parameters used in the model are as follows:

We assume that bacteria follow a logistic growth with carrying capacity $T$. Let $\beta_s$ and $(1-c)\beta_r$ the birth rate of sensitive and resistant bacteria, respectively. Specific mutations arising from resistance to chemical control frequently have an inherent fitness cost which may be resulted through reduced reproductive capacity or competitive ability [1,22,25,45,46-49]. We quantify fitness cost as a reduction on the reproduction rate of the resistant strain, therefore $0 < c < 1$. Also, the immune cells proliferate proportionally (with a proportionality constant $\gamma$) to the bacterial load [42]. This situation can be express a generalised model of a local bacterial infection, such as a urinary tract, tuberculosis or wound infection.

Immune cells, sensitive and resistant bacteria have per capita natural death rates $\mu_B$, $\mu_S$ and $\mu_R$, respectively [1,3,15,43,45]. During the administration of the $i$-th antibiotic, a number of resistant bacteria to it can show up due to mutations of exposed sensitive bacteria to such antibiotic, we model this situation by the term $\overline{\mu_i}A_iS$ where $\overline{\mu_i}$ is the mutation rate of sensitive bacteria due to exposure to $i$-th antibiotic [1]. Sensitive and resistant bacteria have per capita death rates by response of
immune cells and this rates is \( \eta \) [3,15,39,42,43]. We assume that the predation of the \( i \)-th antibiotic on sensitive bacteria follows a Holling function of the 2nd type [3,45]. In this respect, sensitive bacteria also die due to the action of the \( i \)-th antibiotic, and we presume that the effect of the \( i \)-th antibiotic on susceptible bacteria for \( i = 1,2,\ldots,n \) is modelled using a saturating response, \( \frac{E_{\max}A_i}{E_{\max}+A_i} \), subject to a maximum killing rate \( E_{\max} \) and the antibiotic concentration required for half maximum effect, \( E^0_{\max} \) [22,50-52]. Lastly, the \( i \)-th antibiotic concentration is supplied at a constant rate \( \delta_i \), and is taken up at a constant per capita rate \( \mu_i \).

Under the assumptions aforementioned, we obtain the following system of \( (n+3) \) ODE's:

\[
\frac{ds}{dt} = \beta_S(1 - s + r) - \mu_s s - \eta b s - s \sum_{i=1}^{n} a_i \left( \frac{E_{\max}^i}{E_{\max}^i + A_i} \right)
\]

\[
\frac{dr}{dt} = \beta_R(1 - s + r) - \mu_r r - \eta b r + s \sum_{i=1}^{n} a_i A_i
\]

\[
\frac{db}{dt} = \beta(1 - s + r) - \mu b - \beta b r s - s \sum_{i=1}^{n} a_i A_i
\]

\[
\frac{dA_i}{dt} = \delta_i - \mu_i A_i, \quad \text{for} \quad i = 1,2,\ldots,n
\]

where all the parameters, \( \beta_S, \beta_R, T, \mu_s, \mu_r, \mu_b, \eta \) and \( \delta_i, \mu_i, E_{\max}^i, E_{\max}^i \) for \( i = 1,2,\ldots,n \), are positive. To reduce the number of parameters, we changed the variables as follows:

\[
s = \frac{S}{r}, \quad r = \frac{R}{r}, \quad b = B, \quad a_i = \frac{A_i}{\delta_i}
\]

With the new variables, the normalized system is as given:

\[
\frac{ds}{dt} = \beta_S(1 - s + r) - \mu_s s - \eta b s - s \sum_{i=1}^{n} a_i \left( \frac{E_{\max}^i}{E_{\max}^i + A_i} \right)
\]

\[
\frac{dr}{dt} = \beta_R(1 - s + r) - \mu_r r + s \sum_{i=1}^{n} a_i A_i
\]

\[
\frac{db}{dt} = \beta(1 - s + r) - \mu b - \beta b r s - s \sum_{i=1}^{n} a_i A_i
\]

where \( \beta_S = (1 - c) \beta_S, \gamma = \beta T \) and \( a_i = \frac{A_i}{\delta_i} \)

\[
E_{\max}^i = \frac{E_{\max}^i}{\mu_i} \quad \text{for} \quad i = 1,2,\ldots,n
\]

The studied region as biological is given by the set

\[
\Omega = \{ (s, r, b, A_1, A_2, \ldots, A_n) \in \mathbb{R}^{n+3}; 0 \leq s(t) + r(t) \leq 1, 0 \leq b(t), 0 \leq a_i(t), \leq 1 \quad \text{for} \quad i = 1,\ldots,n \}.
\]

The following proposition ensures that system (2) is well posed in the sense that solutions with initial conditions started in \( \Omega^+ \) remain in the region \( \Omega \) for all \( t \geq 0 \), and so, this solutions of system (2) have biological meaning.

**Proposition 2.1** The region \( \Omega \) defined in (3) is positively invariant with respect to the system(2).

**Proof** By adding the first two equations of the system (2).

\[
\frac{ds}{dt} + \frac{dr}{dt} = (\beta_S s + \beta_R r)(1 - s + r) - \mu_s s - \mu_r r - \eta b (s + r) - s \sum_{i=1}^{n} \frac{a_i E_{\max}^i}{E_{\max}^i + A_i} \quad \text{(4)}
\]

is obtained. Considering the region \( \Omega \), we have the following inequality:

\[
\frac{ds}{dt} + \frac{dr}{dt} \leq \beta_S(1 - s + r) \quad \text{(5)}
\]

By the qualitative analysis of inequality (5), it follows that \( 0 \leq s(t) + r(t) \leq 1 \) for all \( t \geq 0 \). Furthermore, the solutions of the last \( n \) equations of system (2) are

\[
a_i(t) = 1 + (-1 + a_i(0)) e^{-\mu_i t}, \quad \text{for} \quad i = 1,2,\ldots,n \quad \text{(6)}
\]

where initial conditions satisfy \( 0 < a_i(0) \leq 1 \) for \( i = 1,2,\ldots,n \). By (6), it is found that \( 0 \leq a_i(t) \leq 1 \). Lastly, let \( 0 \leq s(t) + r(t) = u(t) \leq 1 \), then the solution of third equation in system (2) is

\[
b = b(0) e^{(1-\gamma) b u(t)}, \quad \text{where} \quad 0 < b(0) < b(t) \quad \text{(7)}
\]

In this context, solutions starting in \( \Omega \) remain there for all \( t \geq 0 \).

### 3. Qualitative analysis of system (2)

We have examined the existence and stability of equilibria of system (2).

**Proposition 3.1** Let

\[
\frac{\beta_S - \mu_s}{\beta_R - \mu_r} = A_3, \quad \frac{\beta_S}{\beta_R} = A_4, \quad \frac{\sum_{i=1}^{n} a_i}{\eta} = A_5, \quad \frac{\beta}{\gamma} = A_6
\]

where \( 0 < A_3, A_4, A_5, A_6 \). It is accepted that the general expressions of the system’s equilibria show as

\[
E_i = \left( s, r, b, A_1, \ldots, A_n \right) \quad \text{for} \quad i = 1,2,\ldots,n \quad \text{and} \quad j = 0,1,2,3,4.
\]

Then, the system (2) always has \( E_0 = (0,0,0,1,1,\ldots,1) \) (namely, the infection-free equilibrium point) contained in \( \Omega \). Also, in the region \( \Omega \), there are equilibria that \( E_1 = (0, A_3, 0,1,\ldots,1) \) when \( 0 < A_3 \), \( E_2 = (0, A_6, A_3, A_3, A_3, A_3, 1,\ldots,1) \) when \( A_6 < A_3 \), and

\[
E_3 = \left( A_1 - A_3 + A_3, A_1 - A_3 + A_3, A_1 - A_3 + A_3, A_1 - A_3 + A_3, A_1 - A_3 + A_3, A_1 - A_3 + A_3, 1,\ldots,1 \right)
\]

when \( \max(0,A_3) < A_1 \). In addition to \( E_0 \), \( E_1 \) and \( E_2 \), if \( A_3 - A_4 A_1 - A_3 + A_1, A_1 - A_3 + A_3, \ldots, A_1 - A_3 + A_3 \), then there exists a fifth equilibrium point,

\[
E_4 = \left( A_1 - A_3 + A_3, A_1 - A_3 + A_3, A_1 - A_3 + A_3, A_1 - A_3 + A_3, A_1 - A_3 + A_3, A_1 - A_3 + A_3, 1,\ldots,1 \right), \quad \text{in} \quad \Omega.
\]

**Proof** The equilibria of system (2) are given by the solutions of the system of following algebraic equations;
From the last n equations of system (9), we have 
\( \overline{a}_i = 1 \) for \( i = 1, 2, \ldots, n \). Consequently, the system (9) turns into following system:

\[
\begin{align*}
\beta_x (t - (s + r) - \mu_x - \eta_b s - \sum_{i=1}^n a_i E_x^{i} (c_{i,3} + a_i) &= 0 \\
\beta_y (t - (s + r) - \mu_y - \eta_b y + \sum_{i=1}^n a_i a_i &= 0 \\
\beta_z (t - (s + r) - \mu_z - \eta_b z + \sum_{i=1}^n a_i &= 0 \\
\beta_d (t - (s + r) - \mu_d - \eta_b d + \sum_{i=1}^n a_i &= 0
\end{align*}
\]

Considering the equalities in (8), we have that

\[
\overline{A}(A_1 - (\overline{A} + \overline{F}) - A_2) = 0
\]

\[
\overline{T}(A_3 - (\overline{A} + \overline{F}) - A_4) + A_5 = 0
\]

\[
\overline{B}(\overline{A} + \overline{F} - A_6) = 0
\]

From (11), it is obtained that either \( \overline{F} = 0 \) or \( \overline{F} + \overline{A} = A_4 - A_2 \). When \( \overline{F} = 0 \), the solutions of system (11) are that

\[
E_0 = (0, 0, 0, 1, \ldots, 1), \quad E_1 = (0, A_4, 0, 0, \ldots, 1) \quad \text{and} \quad E_2 = (0, A_6, A_3 - A_6, A_3, 0, 1, \ldots, 1).
\]

Equilibrium point \( E_0 \) always exists in \( \Omega \). Also, the conditions to be biologically meaningful of the equilibrium points \( E_1 \) and \( E_2 \) are

\[
\frac{A_5}{A_4} < A_3 < A_2 \quad \text{and} \quad A_6 < A_3 < A_2,
\]

respectively.

In the same way, if \( \overline{F} + \overline{A} = A_4 - A_2 \), then the solutions are that

\[
E_3 = \left( A_1, A_2 - A_3, A_2 - A_3 + A_4, A_4, A_4, 0, 1, \ldots, 1 \right)
\]

\[
E_4 = \left( A_6, A_6 A_3 - A_4 + A_5, A_6, A_6 A_3 - A_4 + A_5, A_6 A_4 - A_3 + A_5, A_6 A_4 - A_3 + A_5, A_6 A_5 - A_2 + A_5, A_6 A_4 - A_3 + A_5 \right)
\]

As a consequence, the equilibria of system (2) in \( \Omega \) have been summarized in the following table.

| Table 1 Biological existence conditions of the equilibria of system (2). |
|---|---|
| Equilibrium Points | Biological Existence Conditions |
| \( E_0 = (0, 0, 0, 1, \ldots, 1) \) | Always exists |
| \( E_1 = (0, A_4, 0, 0, \ldots, 1) \) | \( 0 < A_3 \) |
| \( E_2 = (0, A_6, A_3 - A_6, A_3, 0, 1, \ldots, 1) \) | \( A_6 < A_3 < A_2 \) |
| \( E_3 = \left( A_1, A_2 - A_3, A_2 - A_3 + A_4, A_4, A_4, 0, 1, \ldots, 1 \right) \) | \( \max(0, A_3) < A_3 \) |
| \( E_4 = \left( A_6, A_6 A_3 - A_4 + A_5, A_6, A_6 A_3 - A_4 + A_5, A_6 A_4 - A_3 + A_5, A_6 A_4 - A_3 + A_5, A_6 A_5 - A_2 + A_5, A_6 A_4 - A_3 + A_5 \right) \) | \( A_1 - A_6 A_4 - A_3 + A_5 < A_6 < A_1 \) |

Theorem 3.1 (Routh-Hurwitz Criteria): Given the polynomial,

\[
P(\lambda) = \lambda^n + a_1 \lambda^{n-1} + \ldots + a_{n-1} \lambda + a_n,
\]

where the coefficients \( a_i \) are real constants, \( i = 1, \ldots, n \), define the \( n \) Hurwitz matrices using the coefficients \( a_i \) of the characteristic polynomial:

\[
H_1 = (a_1), H_2 = \left( a_1 \ a_2 \ 1 \ a_3 \ a_4 \ a_5 \ a_6 \right), H_3 = \left( a_1 \ a_2 \ a_3 \ a_4 \ a_5 \ a_6 \right), \ldots, H_n
\]

where \( a_0 = 0 \) if \( j > n \). All of the roots of polynomial \( P(\lambda) \) are negative or have negative real parts if and only if the determinants of all Hurwitz matrices are positive: \( \det H_j > 0 \), \( j = 1, 2, \ldots, n \). For polynomial of degree \( n = 2, 3, 4 \) and \( 5 \), the Routh-Hurwitz criteria are summarized.

Theorem 3.2 Suppose \( \frac{dX}{dt} = F(X) \) is a nonlinear first-order autonomous system with an equilibrium \( \overline{X} \). Denote the Jacobian matrix of \( F \) evaluated at \( \overline{X} \) as \( J(\overline{X}) \). If the characteristic equation of the Jacobian matrix \( J(\overline{X}) \),

\[
\lambda^n + a_1 \lambda^{n-1} + a_2 \lambda^{n-2} + \ldots + a_{n-1} \lambda + a_n = 0
\]

satisfies the conditions of the Routh-Hurwitz criteria in Theorem 3.1, that is, the determinants of all of the Hurwitz matrices are positive, \( \det(H_j) > 0 \), \( j = 1, 2, \ldots, n \), then the equilibrium \( \overline{X} \) is locally asymptotically stable. If \( \det(H_j) < 0 \), for some \( j = 1, 2, \ldots, n \), then the equilibrium \( \overline{X} \) is unstable [53].

The following proposition has shown conditions that equilibrium points in the proposition 3.1 are locally asymptotically stability (LAS).

Proposition 3.2 The equilibrium points of system (2) in \( \Omega \) satisfy

(i) If \( A_1, A_2 < 0 \), then \( E_0 \) is LAS.

(ii) Let \( 0 < A_3 \). If \( A_1 < A_2 < A_6 \), then \( E_1 \) is LAS.

(iii) Let \( A_6 < A_3 \). If \( A_1 - A_6 - A_2 < A_3 - A_6 < 0 \), then \( E_2 \) is LAS.
(iv) Let $\max\{0, A_3\} < A_1$. If $A_1 < \min\{A_2, A_6\}$, then $E_3$ is LAS.

(v) Let $A_3 - A_4 \frac{A_1 - A_6}{A_2} < A_6 < A_1$. In this case $E_4$ is unstable point.

**Proof** For the stability analysis, the functions of the right side of system (2) are determined as follows;

\[
\begin{align*}
\varphi_1(s, r, b, a) &= \psi(s, r, b, a) - s \beta s - (s + r) \mu - \eta b = 0 \\
\varphi_2(s, r, b, a) &= \psi(s, r, b, a) - s \beta s - (s + r) \mu - \eta b + s \sum a_i a_i \\
\varphi_3(s, r, b, a) &= \psi(s, r, b, a) - b(y(s + r) - \mu)
\end{align*}
\]

\[
(12)
\]

\[
J = \begin{pmatrix}
\beta s - 2\beta s - \beta s \tau + \mu - \eta b \\
-\sum a_i \psi_0 \frac{s \mu_0 + \psi_0}{s \mu_0 + \psi_0} - a_i
\end{pmatrix}
\]

\[
(13)
\]

Since it is found as $a_i = 1$ for $i = 1, 2, \ldots, n$ in the equilibria of system (2), the matrix (13) transforms to

\[
J = \begin{pmatrix}
\beta s (A_1 - 2s - r - A_2 b) & -\beta s \\
\beta s (A_1 - 2s - r - A_2 b) & -\beta s + \eta b \\
0 & \eta b \\
\end{pmatrix}
\]

\[
(14)
\]

by (8).

For ease of examination, the $\tau$-th eigenvalue of equilibrium point $E_k$ was shown as $\lambda_{k, \tau}$ for $k = 0, 1, 2, 3, 4$ and $\tau = 1, 2, \ldots, n$ in $N$.

(i) The jacobian matrix in (14) evaluated at the equilibrium point $E_0$ in $\Omega$ is

\[
J(E_0) = \begin{pmatrix}
\beta s A_1 & 0 & 0 \\
\beta s A_2 & \beta s A_3 & 0 \\
0 & -A_4 s & 0
\end{pmatrix}
\]

\[
(15)
\]

The eigenvalues obtained from (15) are that $\lambda_{0, 0} = \beta s A_1$, $\lambda_{0, 0} = \beta s A_3$, $\lambda_{0, 0} = -\gamma A_6$ and $\lambda_{0, 0} = -\mu_1$ for $i = 1, 2, \ldots, n$. It is clear that $0 < A_6$ due to (8). According to theorem 3.2, if $A_1, A_3 < 0$, the infection-free equilibrium point $E_0$ in $\Omega$ is LAS for system (2).

(ii) Let $0 < A_3$. In this case, there is equilibrium point $E_1$ in $\Omega$. In the same way, the jacobian matrix obtained from the equations in (12) is (13) and, since it is found as $a_i = 1$ for $i = 1, 2, \ldots, n$ in the equilibria of system (2), the matrix (13) transforms to (14).

For ease of examination, the $\tau$-th eigenvalue of equilibrium point $E_k$ was shown as $\lambda_{k, \tau}$ for $k = 0, 1, 2, 3, 4$ and $\tau = 1, 2, \ldots, n$ in $N$. 

There are $E_2$ in $\Omega$. Jocobian matrix evaluated at $E_2$ is as following matrix:

\[
J(E_2) = \begin{pmatrix}
\beta s (A_1 - 2s - r - A_2 b) & -\beta s \\
\beta s (A_1 - 2s - r - A_2 b) & -\beta s + \eta b \\
0 & \eta b \\
\end{pmatrix}
\]

\[
(16)
\]

The eigenvalues of matrix (17) are $\lambda_{2, 1+2} = -\mu_1 < 0$ for $i = 1, 2, \ldots, n$ and the others are found from following matrix;

\[
J(E_3) = \begin{pmatrix}
\beta s (A_1 - 2s - r - A_2 b) & 0 & 0 \\
\beta s (A_1 - 2s - r - A_2 b) & -\beta s & -\eta b - \mu_1 \\
0 & 0 & 0
\end{pmatrix}
\]

\[
(17)
\]
where matrix $J^B(E_2)$ is the block matrix of (17). Characteristic equation of (18) is

$$J^B(E_2) = \begin{pmatrix} \lambda^2 + \beta_2 \lambda + \eta \gamma & \frac{A_2 - A_6}{A_4} \\ \frac{A_2 - A_6}{A_4} & -\eta \gamma \end{pmatrix}$$

Then, the remain three eigenvalues are determined by solving of equation (19).

Hence, the first eigenvalue is

$$\lambda_{2,1} = \beta_2 \left( A_1 - A_6 - A_2 \frac{A_3 - A_6}{A_4} \right)$$

and the other two eigenvalues are gained by solving following equation:

$$\lambda^2 + \beta_2 \lambda + \eta \gamma \frac{A_3 - A_6}{A_4} = 0$$

In the equation (21), it is clear that

$$A_6 \frac{A_3 - A_6}{A_4} > 0$$

due to the (16). Thus, all Hurwitz matrices are positive from theorem 3.1 ($n = 2$). In this respect, it is obtained that $Re \lambda_{2,2}, Re \lambda_{2,3} < 0$ for equation (21). According to theorem 3.2, the conditions for LAS are provided for all eigenvalues except $\lambda_{2,3}$ given in the (20). It is sufficient to examine the sign of $\lambda_{2,3}$. Accordingly, it is obtained that $A_1 - A_6 - A_2 \frac{A_3 - A_6}{A_4} < 0$.

From (16), If $A_1 - A_6 - A_2 \frac{A_3 - A_6}{A_4} < 0$ and $A_6 < A_3$, then equilibrium point $E_2$ in $\Omega$ is LAS for system (2).

(iv) Let

$$\max(0, A_3) < A_1.$$ (22)

In this case, equilibrium point $E_2$ reveals in $\Omega$. The eigenvalues of jacobian matrix evaluated at $E_2$ is

$$\lambda_{3,3} = \gamma(A_1 - A_2)$$ and $\lambda_{i,3} = -\mu_1$ for $i = 1,2,\ldots,n$. (23) and the others are found from following matrix;

$$J^B(E_2) = \begin{pmatrix} \beta_2 A_1 (A_1 - A_2) & \beta_2 A_1 (A_1 - A_2) \\ \frac{A_1 - A_2}{A_4} & -\eta \gamma \end{pmatrix}$$

when matrix $J^B(E_2)$ is considered as the block matrix of (23). Meanwhile, when

$$A_1 < A_6,$$ (25) we have that $Re \lambda_{3,3} < 0$. Trace and determinant of matrix (24) for $E_2$ are

$$\text{Tr}J^B(E_2) = -\beta_2 A_1 (A_1 - A_2) + \mu_1 \left( A_1 - A_2 + \frac{A_3 - A_6}{A_4} \right)$$

and

$$\text{Det}J^B(E_2) = -\beta_2 A_1 (A_1 - A_2),$$

respectively. We have that $\text{Det}J^B(E_2) > 0$ and $\text{Tr}J^B(E_2) < 0$ due to (22), and so,

$$Re \lambda_{3,3,1} < 0.$$ By (22) and (25), if

$$\max(0, A_3) < A_1 < A_6$$ (27) then, $E_2$ is LAS from theorem 3.2.

(v) Lastly, let $A_1 - A_4 \frac{A_1 - A_2}{A_2} < A_6 < A_1$. Then the equilibrium point $E_4$ reveals in $\Omega$. Jacobian matrix in (14) evaluated at $E_4$ is

$$J^B(E_4) = \begin{pmatrix} \frac{A_1 - A_2}{-\tau - \Delta_4} & -\mu_2 & -\eta \gamma & \tau \left( \frac{E_{1,2} E_{2,2}}{1 + \sigma_1} \right) & \cdots & \tau \left( \frac{E_{n,1} E_{1,1}}{1 + \sigma_1} \right) \\ \beta_2 \left( A_1 - A_6 \right) & -\eta \gamma & \tau \left( \frac{E_{1,2} E_{2,2}}{1 + \sigma_1} \right) & \cdots & \tau \left( \frac{E_{n,1} E_{1,1}}{1 + \sigma_1} \right) \\ \frac{A_1 - A_2}{-\tau - \Delta_4} & -\eta \gamma & \tau E_{1,1} & \cdots & \tau E_{1,n} \\ \vdots & \vdots & \vdots & \ddots & \vdots \\ \frac{A_1 - A_2}{-\tau - \Delta_4} & \frac{A_1 - A_2}{-\tau - \Delta_4} & \frac{A_1 - A_2}{-\tau - \Delta_4} & \cdots & \frac{A_1 - A_2}{-\tau - \Delta_4} \end{pmatrix}$$

where

$$E_4 = \left( \begin{array}{cc} A_6 - A_2 - A_4 & A_6 - A_2 - A_4 \\ \frac{A_6 - A_2 - A_4}{A_5} & A_6 - A_2 - A_4 \end{array} \right),$$

when matrix $J^B(E_4)$ is considered as the block matrix of (28). Characteristic equation of matrix (29) is obtained as follows:

$$\lambda^2 + \alpha_1 \lambda^2 + \alpha_2 \lambda + \alpha_3 = 0.$$ (30)

where

$$a_1 = \frac{\tau}{\tau A_2 + \tau} \beta_4 + \beta_5 \bar{b},$$

$$a_2 = \frac{\tau}{\bar{b} + \tau} \beta_4 \beta_5 \bar{b} - \beta_4 A_2 \bar{b} - \beta_5 \bar{b} \right) - \beta_2 A_2 \bar{b} \bar{b}$$

and

$$a_3 = -\beta_2 A_2 \bar{b} A_2 \bar{b} \left( 1 + \frac{\tau}{\tau} \right) < 0.$$ (31)

From theorem 3.1 ($n = 3$), the conditions for $Re \lambda_{3,4}, A_1, A_3 < 0$ are $a_1, a_2 > 0$ and $a_1 a_2 - a_3 > 0$. Since $a_3 < 0$ in (31), the equilibrium point $E_3$ in $\Omega$ is unstable point for the system (2). Hence, proof is completed.

For equilibriums of system (2) in $\Omega$, the conditions found for LAS and biological existence are summarized in the table below.
Table 2 The biological existence and LAS conditions of the equilibria of system (2).

<table>
<thead>
<tr>
<th>Equilibrium Points</th>
<th>Biological Existence and LAS Conditions</th>
</tr>
</thead>
<tbody>
<tr>
<td>$E_0 = (0,0,1,0,0)$</td>
<td>$A_0, A_1 &lt; 0$</td>
</tr>
<tr>
<td>$E_1 = (0, A_0, 0, 1, 0)$</td>
<td>$\max(0, A_0) &lt; A_1 &lt; A_0$</td>
</tr>
<tr>
<td>$E_2 = (A_0, A_0, A_0, A_0, 1)$</td>
<td>$A_0 &lt; A_1$ and $A_0 - A_1 &lt; A_0$</td>
</tr>
<tr>
<td>$E_3 = (A_0, A_0, A_1, A_1, 1)$</td>
<td>$\max(0, A_0) &lt; A_1 &lt; A_0$</td>
</tr>
</tbody>
</table>

Proposition 3.3 Let us denote by $\Omega_0$ the region where the equilibrium point $E_0$ in $\Omega$ exists biological and is LAS. In the same way, $\Omega_1$, $\Omega_2$ and $\Omega_3$ is for $E_1$, $E_2$ and $E_3$, respectively. Then $\Omega_i \cap \Omega_j = \emptyset$ for $i$, $j = 0, 1, 2, 3$ and $i \neq j$.

**Proof** It can be clearly seen in table 2.

Proposition 3.4 Let $E_1$ is LAS. Then the equilibrium point $E_1$ in $\Omega$ is globally asymptotically stable (GAS) for system (2).

**Proof** In the system (2), the variable $r$ has logistic form. For $E_1$, it is examined as follows: we have the region $\Omega_1 = \{(r, s) \in \mathbb{R}^2 : 0 \leq r \leq 1\}$ given by

$$dr \quad dt = \beta_r r(1-r) - \mu_r r, \quad (32)$$

where $\beta_r > 0$ is the intrinsic growth rate, $\mu_r > 0$ is the natural death rate and $F_1 = 1$ is the carrying capacity. Considering (8), there are two equilibria $\Omega_i = \{(r, s) \in \mathbb{R}^2 : \text{in this respect, } r(t) \leq A_1\}$. When the differential equation in (32) can be solved by separation of variables, it is obtained that $r(t) = A_3 \frac{r(0)}{r(0) + e^{-\lambda t}}$. It can be seen $\lim_{t \to \infty} r(t) = A_3$. Thus, $F_1 = A_3$ (that is, $E_1$) is GAS.

Proposition 3.5 Let $E_2$ is LAS. Then, the equilibrium point $E_2$ in $\Omega$ is GAS for system (2).

**Proof** We acquire that the last $n$ equations of system (2) are separated, and their solutions approach $a_i = 1$ for $i = 1, 2, \ldots, n$. Replacing $s = 0$ and $a_i = 1$ for $i = 1, 2, \ldots, n$ in system (2), we attain the asymptotically equivalent planar system [1] in the region $\Omega_2 = \{(r, b) \in \mathbb{R}^2 : 0 \leq r \leq 1, 0 \leq b \}$ given by

$$\begin{align*}
\frac{dr}{dt} &= r(\beta_r (1-r) - \mu_r - \eta b) \\
\frac{db}{dt} &= b(\gamma b - \mu_b)
\end{align*} \quad (33)$$

According to the Dulac criterion, there exists a continuously differentiable function $\Phi_1(r, b)$ for a simply connected region $\Omega_2 \subset \mathbb{R}^2$ such that

$$\Phi_1(r, b) = \frac{1}{rb} \quad (34)$$

Since $(f(r, b), g(r, b))$ is the vector field of system (33),

$$\begin{align*}
\frac{\partial [\Phi_1(r, b)f(r, b)]}{\partial r} + \frac{\partial [\Phi_1(r, b)g(r, b)]}{\partial b} &= \frac{\partial}{\partial r} \left[ \beta_r (1-r) - \mu_r - \eta b \right] \\
&= \frac{\beta_r (1-r) - \mu_r - \eta b}{b} \\
&= -\frac{\beta_r (1-r) - \mu_r - \eta b}{b} < 0.
\end{align*}$$

This result say that system (33) has no periodic orbits contained in the interior of $\Omega_2$ in compliance with Dulac-Bendixon criterion. In addition that, the region $\Omega_2$ cited in the proposition 3.3 does not include another equilibrium point being LAS. Thus, by the Poincaré–Bendixon Theorem and Dulac-Bendixon criterion, we have that equilibrium point $E_2 = \left(0, A_0, \frac{A_0 - A_0}{A_0}, 1, \ldots, 1\right)$ is GAS in $\Omega_2$.

**Proposition 3.6** Let $E_3$ is LAS. In this case, the equilibrium point $E_3$ in $\Omega$ is GAS for system (2).

**Proof** We have that in the same way in the proposition 3.5. Since $E_3$ presents in $\mathbb{R}^2$, we have benefit from Bendixon-Dulac criteria. Let us examine the $E_3$ in the region $\Omega_3 = \{(s, r) \in \mathbb{R}^2 : 0 \leq s + r \leq 1, \beta_2, s, r\}$.

Let $\Phi_2(s, r) = \frac{1}{s}$. Obviously $\Phi_2(s, r) > 0$; functions $F_1(s, r)$ and $F_2(s, r)$ obtained from system (2) are denote as

$$\begin{align*}
F_1(s, r) &= \beta_2 s (1 - (s + r)) - \mu_2 s - \frac{\sum_{i=1}^{n} (E_{s}^{e})}{E_{s}^{e} + 1} - \alpha_i \\
F_2(s, r) &= \beta_2 s (1 - (s + r)) - \mu_2 r + \frac{\sum_{i=1}^{n} \alpha_i}{r} \quad (35)
\end{align*}$$

Then

$$\Delta(s, r) = \frac{\partial}{\partial s} (F_1 \Phi_2) + \frac{\partial}{\partial r} (F_2 \Phi_2).$$

Therefore,

$$\begin{align*}
\Delta(s, r) &= \beta_2 s (1 - (s + r)) - \mu_2 s - \frac{\sum_{i=1}^{n} \alpha_i}{r} \\
&= \beta_2 s (1 - (s + r)) - \mu_2 s - \frac{\sum_{i=1}^{n} \alpha_i}{r} + \frac{1}{r} \left( \frac{E_{s}^{e}}{E_{s}^{e} + 1} - \alpha_i \right)
\end{align*} \quad (36)$$

Thus, by the Bendixon-Dulac criteria, there will be no periodic orbit in the $s - r$ plane. Since $E_3$ is LAS in the above plane and in this region, there is no another equilibrium point being LAS. Therefore, it can be seen that $E_3$ is GAS. Proposition is proved.
4. Numerical study

Among the treatment regimen recommended by WHO includes isoniazid (INH), rifampicin (RIF), streptomycin (SRT) and pyrazinamide (PZA) for some bacterial infections caused by bacteria such as mycobacterium tuberculosis [54]. In this respect, the aforementioned bacteria and antibiotics was used in our numerical study. The values obtained from table 3 are that $A_1 = -0.88812$, $A_2 = 0.375$, $A_3 = 0.22$, $A_4 = 0.75$, $A_5 = 0.00021$, $A_6 = 0.1512$. Also, the GAS due to $A_6 < A_3$ and $A_4 - A_6 - A_2 \frac{A_3-A_5}{A_4} = -1.13105 < 0$.

Table 3: Interpretation and considered values of the parameters. Datas are deduced from the literature.

<table>
<thead>
<tr>
<th>Para.</th>
<th>Description</th>
<th>Unit</th>
<th>Value</th>
<th>Ref.</th>
</tr>
</thead>
<tbody>
<tr>
<td>$\beta_s$</td>
<td>Growth rate of sensitive bacteria</td>
<td>day$^{-1}$</td>
<td>0.8</td>
<td>[1]</td>
</tr>
<tr>
<td>$(1 - c)\beta_s$</td>
<td>Growth rate of resistant bacteria</td>
<td>day$^{-1}$</td>
<td>0.4</td>
<td>[1]</td>
</tr>
<tr>
<td>$\theta &lt; 1$</td>
<td>Growth rate of immune cells as proportionally to the presented bacteria load</td>
<td>day$^{-1}$</td>
<td>$10^9$</td>
<td>[42]</td>
</tr>
<tr>
<td>$\mu_s$</td>
<td>Natural death rate of sensitive bacteria</td>
<td>day$^{-1}$</td>
<td>0.312</td>
<td>[55]</td>
</tr>
<tr>
<td>$\mu_r$</td>
<td>Natural death rate of resistant bacteria</td>
<td>day$^{-1}$</td>
<td>0.312</td>
<td>[56]</td>
</tr>
<tr>
<td>$\mu_b$</td>
<td>Natural death rate of immune cells</td>
<td>day$^{-1}$</td>
<td>1.512</td>
<td>[57]</td>
</tr>
<tr>
<td>$\eta$</td>
<td>Rate of bacteria destroyed by immune cells</td>
<td>cells$^{-1}$</td>
<td>0.3</td>
<td>[45]</td>
</tr>
<tr>
<td>$T$</td>
<td>Carrying capacity of bacteria</td>
<td>bacteria</td>
<td>$10^9$</td>
<td>[25]</td>
</tr>
<tr>
<td>$\sigma_1$</td>
<td>Mutation rate of INH</td>
<td>mut$\times$gen</td>
<td>$10^{-6}$</td>
<td>[54]</td>
</tr>
<tr>
<td>$\sigma_2$</td>
<td>Mutation rate of RIF</td>
<td>mut$\times$gen</td>
<td>$10^{-8}$</td>
<td>[54]</td>
</tr>
<tr>
<td>$\sigma_3$</td>
<td>Mutation rate of SRT</td>
<td>mut$\times$gen</td>
<td>0</td>
<td>[1]</td>
</tr>
<tr>
<td>$\sigma_4$</td>
<td>Mutation rate of PZA</td>
<td>mut$\times$gen</td>
<td>0</td>
<td>[1]</td>
</tr>
<tr>
<td>$E_{i_{max}}$</td>
<td>Maximum killing rate of susceptible bacteria due antibiotic for $i \in 1,2,\ldots,n$ (the average value)</td>
<td>day$^{-1}$</td>
<td>26.4</td>
<td>[58]</td>
</tr>
<tr>
<td>$E_{i_{so}}$</td>
<td>Antibiotic concentration for half maximum effect on susceptible bacteria due antibiotic for $i \in 1,2,\ldots,n$ (the average value)</td>
<td>$\mu g/ml$</td>
<td>0.25-5</td>
<td>[58]</td>
</tr>
<tr>
<td>$\delta_1$</td>
<td>Daily dose of INH</td>
<td>mg/kg/da</td>
<td>5</td>
<td>[54]</td>
</tr>
<tr>
<td>$\delta_2$</td>
<td>Daily dose of RIF</td>
<td>mg/kg/da</td>
<td>10</td>
<td>[54]</td>
</tr>
<tr>
<td>$\delta_3$</td>
<td>Daily dose of SRT</td>
<td>mg/kg/da</td>
<td>15-25</td>
<td>[54]</td>
</tr>
<tr>
<td>$\delta_4$</td>
<td>Daily dose of PZA</td>
<td>mg/kg/da</td>
<td>20-35</td>
<td>[54]</td>
</tr>
<tr>
<td>$\mu_1$</td>
<td>Uptake rate of INH</td>
<td>day$^{-1}$</td>
<td>0.06</td>
<td>[59]</td>
</tr>
<tr>
<td>$\mu_2$</td>
<td>Uptake rate of RIF</td>
<td>day$^{-1}$</td>
<td>0.05</td>
<td>[59]</td>
</tr>
<tr>
<td>$\mu_3$</td>
<td>Uptake rate of SRT</td>
<td>day$^{-1}$</td>
<td>0.04</td>
<td>[59]</td>
</tr>
<tr>
<td>$\mu_4$</td>
<td>Uptake rate of PZA</td>
<td>day$^{-1}$</td>
<td>0.03</td>
<td>[59]</td>
</tr>
</tbody>
</table>

Therefore, qualitative analysis of the system (2) has supported by numerical simulations in the following figure 1 and 2;

Figure 1 Time-dependent changes of sensitive bacteria and antibiotic concentrations by datas obtained from the table 3.

Figure 2 Time-dependent changes of bacteria and immune cells by datas obtained from the table 3.

5. Conclusions

In this paper, we formulated a mathematical model of bacterial resistance to immune system response and multiple antibiotics simultaneously, considering specific changes in bacterial DNA sequence as the only mechanism of bacterial resistance acquisition in order to evaluate the effectiveness of antibiotic treatments with respect to the mechanism above. As mentioned above, values of $A_1, A_2, A_3, A_4, A_5$ and $A_6$ are express the conditions determining the changes in populations of the bacteria, immune cells and concentration of multiple antibiotic. In view of the biological meaning of the parameters defining these expressions, the value $A_3$ is interpreted as the number of bacteria generated by the fraction of sensitive bacteria surviving as a result of its natural death under pressure of multiple antibiotic independently from the effect of immune cells. Analogously, $A_3$ represents the bacteria generated by resistant bacteria surviving as a result of its natural death independently from the effect of immune cells. In this context, the model suggests that if sensitive bacteria can infect and the stream of resistant bacteria reveals in host due effect of
antibiotic, but these do not produce sufficient progeny (in case of $A_2, A_3 < 0$), then these can be disappeared without requiring the action of immune cells (namely, $E_3 = (0,0,0,1,\ldots,1)$ and so, infection is removed.

The value $A_4$ can be expressed that total extinction ratio of immune system cells in the presence of bacteria. If sensitive bacteria can infect and do not produce sufficient progeny, the stream of resistant bacteria reveals in host due effect of antibiotic and can produce sufficient progeny, and total extinction ratio of immune system cells is high enough (in case of $\max\{0,A_3\} < A_4 < A_6$), then immune cells have anymore lost all of activity to infection and so, infection is persistence (namely, $E_3 = (0,A_6 \frac{A_3-A_4}{A_4},1,\ldots,1)$). Another endemic equilibria resembling to this case is for $E_2 = (0,0,1,\ldots,1)$, but the discrepancy in $E_2$ is that total extinction ratio of immune system cells is low enough.

The values $A_2$ and $A_4$ are described as the efficacy of the immune system on the sensitive and resistant bacteria, respectively. When sensitive bacteria can infect, the stream of resistant bacteria reveals in host due effect of antibiotic, these can produce sufficient progeny, and total extinction ratio of immune system cells is high enough (in case of $\max\{0,A_3\} < A_4 < A_6$), then immune cells have anymore lost all of activity to infection and so, infection is persistence (namely, $E_3 = (0,A_6 \frac{A_3-A_4}{A_4},1,\ldots,1)$). Our model is quite appropriate when compared to the complexity of biological phenomenon and it predicts in terms of these values when the bacterial progression is either for resistant bacteria as shown in figure 1 and 2 or for sensitive and resistant bacteria. Moreover, it estimates the situation where there is no bacterial progress too.

The $E_0$ is a condition that affects only the appropriate use of antibiotics. For $E_1$ and $E_3$, these results in the our model highlight the fact that those whose immunity response against infections have diminished, suffer from the same bacterial infections more. Furthermore, this model shows that some of the bacterial infections believed its have limited or destroyed, make an individual whose immune system deteriorated suffer more. In this respect, the effects of antibiotics are much than assumed, since these are used probable inappropriately or random. Thus, the appropriate dose and duration of antibiotics play the major role in these infections. In this case, the effect and magnitude of immune cells disappear by gradually decreasing for these infections.

For $E_2$, in the individuals who receive not in the appropriate dose and duration of antibiotic cocktail according to the type and characteristic of the bacteria causing infection, infection is limited but persistence. It continues only for the resistant population with host's immune cells, it is necessary to develop the treatment strategies for the different antibiotic and to strengthen the immune system. In this case, when an individual's immune system weakens, he/she suffers more from the bacterial infections such as tuberculosis which are believed to have been confined or terminated.

Additionally, the results obtained from numerical studies in terms of bacterial infection reveal the affinity substantially with reference to the clinical treatment.

For future work we are planing consider other mechanisms such as the loss of resistance in the resistant bacteria and gaining resistance by conjugating of sensitive and resistant bacteria in order to get more accurate results.


