Research Article

Dynamics of Immune Checkpoints, Immune System, and BCG in the Treatment of Superficial Bladder Cancer

Farouk Tijjani Saad, Evren Hincal, and Bilgen Kaymakamzade

Department of Mathematics, Near East University, North Nicosia, Northern Cyprus, Mersin 10, Turkey

Correspondence should be addressed to Farouk Tijjani Saad; farouksaaad@yahoo.co.uk

This paper aims to study the dynamics of immune suppressors/checkpoints, immune system, and BCG in the treatment of superficial bladder cancer. Programmed cell death protein-1 (PD-1), cytotoxic T-lymphocyte-associated antigen 4 (CTLA4), and transforming growth factor-beta (TGF-β) are some of the examples of immune suppressors/checkpoints. They are responsible for deactivating the immune system and enhancing immunological tolerance. Moreover, they categorically downregulate and suppress the immune system by preventing and blocking the activation of T-cells, which in turn decreases autoimmunity and enhances self-tolerance. In cancer immunotherapy, the immune checkpoints/suppressors prevent and block the immune cells from attacking, spreading, and killing the cancer cells, which leads to cancer growth and development. We formulate a mathematical model that studies three possible dynamics of the treatment and establish the effects of the immune checkpoints on the immune system and the treatment at large. Although the effect cannot be seen explicitly in the analysis of the model, we show it by numerical simulations.

1. Introduction

Cancer is a class of diseases characterized by out-of-control cell growth which affects and damages the DNA. Cancer prevalence is increasing in many countries [1]. Many treatment options of cancer exist, which include surgery, immunotherapy, chemotherapy, radiotherapy, vaccine therapy, and hormonal therapy [1, 2]. The mode and type of treatment depend on the type, location, and grade of the cancer and the patient's body. The bladder is a hollow organ in the lower abdomen which collects urine produced by the kidneys. Bladder cancer is a growth of malignant cells initiating in the urinary bladder. It is common, with around 38,000 men and 15,000 women diagnosed every year in the United States. Approximately 400,000 new cases are diagnosed and about 150,000 die directly from the disease every year across the globe [3, 4].

The bladder wall is lined with cells called transitional and squamous cells. The most common type of bladder cancer is urothelial carcinoma or transitional cell carcinoma (TCC). It mostly originates from the transitional cells and further progresses and grows superficially on the inner surface of the bladder; as a result, it invades the bladder wall and vessels, dispersing into the neighboring organs as well as forming distant metastases [5–7].

One of the most effective ways of treating bladder cancer is immunotherapy. This is the process of stimulating, activating, and triggering the immune system to spread, locate, and kill cancer cells [8].

Intravesical Bacillus Calmette-Guerin (BCG) is an attenuated nonpathogenic strain of *Mycobacterium bovis* that was initially used as a vaccine against tuberculosis. The attenuation was reached via manipulation of the bacillus by serial growth on a culture medium. As a result, the genes causing virulence will be lost and inoculated into humans [9, 10]. It is undoubtedly the most efficient and successful immunotherapy of cancer [9]. BCG therapy is used for various types of cancers, including acute lymphoblastic leukemia and melanoma. The first report of successful use of BCG to treat patients with bladder cancer was in 1976 by Morales et al. They obtained the efficacy of BCG therapy and established it as the pillar for the treatment of non-muscle-invasive bladder cancer after transurethral resection [5, 11].

Intravesical BCG is a type of immunotherapy that is also used to treat superficial bladder cancer [12, 13]. It is usually applied after local surgery to prevent tumor recurrence. It is given in 6-weekly intravesical instillation of 1.5×10^8 bacteria, which has been proven to be superior to chemotherapy in reducing recurrence rates of the tumor [12–14]. When the BCG is instilled and processed into the bladder, it creates an inflammatory environment which in turn stimulates an immune response, resulting in attacking the cancer cells. Therefore, many researchers believed that BCG reduces tumor progression and stated that the primary role of BCG treatment is to stimulate, trigger, and activate the immune effector cells in order to attack the cancer cells. In spite of the fact that BCG instillation is regarded as the "gold standard" treatment, it has many side effects which include hematuria, pain, dysuria, and fever, to mention a few [7–14].

Immune checkpoints are negative regulators of the immune system which play important roles in maintaining self-tolerance, preventing autoimmunity, and protecting tissues from immune collateral damage. These immune checkpoints are often hijacked by tumors to restrain the ability of the immune system to mount an effective antitumor response. The tumors neutralize some immune checkpoint pathways in order to maintain immune resistance, particularly against T-cells. The T-cells are specific tumor antigens. Examples of the aforementioned checkpoints are PD-1 and CTLA4 [15–17].

Programmed cell death protein-1 (PD-1) is a protein that is encoded by the PDCD1 gene in humans. It is a cell surface receptor which belongs to the immunoglobulin superfamily and is expressed on T-cells and pro-B-cells. PD-1 binds two ligands, PD-L1 and PD-L2. The PD-1 acts as an immune checkpoint, which plays an important role in downregulating the immune system by preventing the activation of the T-cells. Hence, it decreases autoimmunity and encourages self-tolerance [18, 19]. The immune system is directly affected by the activities of PD-1 in the sense that it suppresses, blocks, and deactivates the immune cells from spreading, fighting, and attacking the cancer cells. Therefore, PD-1 aids in growth, development, and progression of the cancer. In conclusion, it disrupts and affects immunotherapy [20–24].

Transforming growth factor-beta 1 (TGF-β1) is a regulatory cytokine which suppresses immune function in cancers and in chronic viral infections. It inhibits the activation of the T-cells and subdues their proliferation. Hence, cancer cells take advantage of this immune checkpoint pathway as a way to escape and evade detection. This leads to the inhibition of antitumor immune response, resulting in cancer growth and development [25, 26].

Mathematical modeling and simulation helps in predicting treatments' outcome, as well as describing the behavior and complex dynamics involved. Bunimovich-Mendrazitsky et al. (2007, 2008, and 2011) modeled mathematically the use of BCG in noninvasive bladder cancer, where their study identified fixed points and conditions for stability of the dynamical system [6, 8, 14]. In 2016, Bunimovich-Mendrazitsky developed a new mathematical model for combined BCG and IL-2 bladder cancer treatment which

introduces the effect of TAA T-cells. Furthermore, Starkov utilized a mathematical approach for bladder cancer treatment model in the derivation of ultimate upper and lower bounds. He also presented tumor clearance conditions for BCG treatment of bladder cancer [13].

In this research, we formulate a mathematical model to study the dynamics of immune checkpoints/suppressors, immune system, and the BCG immunotherapy of bladder cancer. Moreover, we highlight the effects of immune checkpoints/suppressors on the immune system and the treatment numerically.

This paper is organized as follows. Section 1 is the introduction. Section 2 is the formulation and presentation of our model. We give the stability analysis and numerical simulations in Sections 3 and 4, respectively. In the final section, we state our conclusions and discussions.

2. Formulation of the Model

The model consists of a system of four nonlinear differential equations, which characterize the dynamics of the interaction between cancer cells (C), different arms of the immune system regarded as effector cells (E), the BCG (B), and all categories of immune suppressors/checkpoints as (P).

2.1. Dynamics of Cancer Cells. The dynamics of cancer cells is given by

$$\frac{dC}{dt} = rC - \frac{\alpha_1 EC}{P + k}. \tag{1}$$

Here, we assume that, in the absence of the immune system, the cancer cells grow exponentially with growth rate r. The second term shows the elimination of cancer cells by the effector cells at the rate α_1, while $1/(P + k)$ is the immunosuppressive factor by the immune checkpoints/suppressors, which interrupts the activities of the effector cells, with k being an inhibitory parameter.

2.2. Dynamics of the Effector Cells. The dynamics of the effector cells is given by

$$\frac{dE}{dt} = \frac{a_1 CE}{P + k} + \frac{a_2 BE}{P + k} - \alpha_2 EC - \mu_1 E. \tag{2}$$

The first term here gives the recruitment of effector cells at the rate a_1 which is directly proportional to the population of cancer cells (i.e., occurring due to the direct presence of cancer cells). $a_2 BE$ shows the activation of effector cells by BCG at the rate a_2. a_1 is the antigenicity of cancer cells which triggers an immune response in the host. It is believed that the immune checkpoints will distort both the recruitment and the activation of effector cells; hence, $1/(P + k)$ is the immunosuppressive response which puts a limitation on the recruitment level and interrupts the activation of effector cells, with k here being an inhibitory parameter. The next term gives the elimination of effector cells by the cancer cells at the rate α_2, and the last term describes the degradation of effector cells at the rate μ_1.

2.3. Dynamics of BCG. The dynamics of BCG is given by

$$\frac{dB}{dt} = b - \alpha_3 EB - \mu_2 B. \tag{3}$$

The first term b is the constant rate of introduction of BCG into the bladder, the second term describes the elimination of BCG by effector cells at the rate α_3, and the third term gives the decay of BCG at the rate μ_2.

2.4. Dynamics of Immune Suppressors/Checkpoints. The dynamics of the immune checkpoints is given by

$$\frac{dP}{dt} = \delta - \mu_3 P. \tag{4}$$

The first term gives the source of immune checkpoints at a constant rate δ, and the second term is the degradation of the immune checkpoints at the rate μ_3.

Finally, the interactions of the cancer cells, effector cells, BCG, and immune checkpoints together lead to the following nonlinear ordinary differential equations:

$$\frac{dC}{dt} = rC - \frac{\alpha_1 EC}{P + k},$$

$$\frac{dE}{dt} = \frac{a_1 CE}{P + k} + \frac{a_2 BE}{P + k} - \alpha_2 EC - \mu_1 E,$$

$$\frac{dB}{dt} = b - \alpha_3 EB - \mu_2 B, \tag{5}$$

$$\frac{dP}{dt} = \delta - \mu_3 P,$$

with initial conditions $C(0) = C_0 \geq 0$, $E(0) = E_0 \geq 0$, $B(0) = B_0 \geq 0$, and $P(0) = P_0 \geq 0$.

2.5. Invariance of Positive Orthant. We show that the system is positively invariant.

From the system, assume $C(0) > 0$, $E(0) > 0$, $B(0) > 0$, and $P(0) > 0$.

From $dC/dt = rC - \alpha_1 EC/(P+k)$, the solution is given by $C(t) = C_0 \exp(\int_0^t (r - \alpha_1 E/(P + k))dt)$. This implies $C(t) > 0$ given that $C_0 > 0$. Also, from $dB/dt = b - \alpha_3 EB - \mu_2 B$, if $B = 0$, then $dB/dt = b > 0$. Therefore, $B(t) > 0$ $\forall t$ since $B_0 > 0$. Moreover, if $b = 0$, then $B(t) = B_0 \exp(-\int_0^t (\alpha_3 E + \mu_2)dt)$

implying $B(t) > 0$ $\forall t$ since $B_0 > 0$. Using $dP/dt = \delta - \mu_3 P$, if $\delta = 0$, *then* $P(t) = P_0 \exp(-\int_0^t \mu_3 dt) > 0$ since $P_0 > 0$.

Also, if $P = 0$ and $\delta > 0$, *then* $dP/dt = \delta$ *which implies* $P(t) > 0$ $\forall t$ *given that* $P_0 > 0$. Now consider $dE/dt = (a_1 C + a_2 B)E/(P+k) - \alpha_2 EC - \mu_1 E$, $E(t) = E_0 \exp(\int_0^t ((a_1 C + a_2 B)/(P + k) - \alpha_2 C - \mu_1)dt) > 0$ given that $E_0 > 0$.

This implies that $E(t) > 0$ $\forall t$ if $E_0 > 0$. Hence, the positive orthant R_+^4 is invariant and $C(t) > 0$, $E(t) > 0$, $B(t) > 0$, and $P(t) > 0$ $\forall t$.

3. Equilibrium and Stability Analysis

3.1. Model without Treatment ($b = 0$). We first analyze our model in the absence of treatment ($b = 0$):

$$\frac{dC}{dt} = rC - \frac{\alpha_1 EC}{P + k},$$

$$\frac{dE}{dt} = \frac{a_1 CE}{P + k} + \frac{a_2 BE}{P + k} - \alpha_2 EC - \mu_1 E,$$

$$\frac{dB}{dt} = -\alpha_3 EB - \mu_2 B, \tag{6}$$

$$\frac{dP}{dt} = \delta - \mu_3 P.$$

The equilibrium points of the model are obtained by equating the equations in (6) to zero and solving simultaneously for C, E, B, and P. They are as follows:

$$U_0 = \left\{ 0, 0, 0, \frac{\delta}{\mu_3} \right\},$$

$$U_1 = \left\{ 0, -\frac{\mu_2}{\alpha_3}, \frac{\mu_1 (\delta + k\mu_3)}{a_2 \mu_3}, \frac{\delta}{\mu_3} \right\}, \tag{7}$$

$$U_2 = \left\{ \frac{\mu_1 (\delta + k\mu_3)}{a_1 \mu_3 - \alpha_2 (\delta + k\mu_3)}, \frac{r (\delta + k\mu_3)}{\alpha_1 \alpha_3}, 0, \frac{\delta}{\mu_3} \right\}.$$

From the invariance of the positive orthant, we concentrate only on the nonnegative equilibria assuming all initial conditions are positive.

As a result, the equilibrium point U_1 will not be considered. Moreover, U_2 exists only if the following condition is satisfied:

$$a_1 \mu_3 > \alpha_2 (\delta + k\mu_3). \tag{8}$$

The Jacobian matrix obtained from (6) is given by

$$\hat{J}(C^*, E^*, B^*, P^*) = \begin{bmatrix} r - \dfrac{\alpha_1 E^*}{P^* + k} & \dfrac{-\alpha_1 C^*}{P^* + k} & 0 & \dfrac{\alpha_1 E^* C^*}{(P^* + k)^2} \\ \dfrac{a_1 E^*}{P^* + k} - \alpha_2 E^* & \dfrac{a_1 C^* + a_2 B^*}{P^* + k} - \alpha_2 C^* - \mu_1 & \dfrac{a_2 E^*}{P^* + k} & \dfrac{-(a_1 C^* E^* + a_2 B^* E^*)}{(P^* + k)^2} \\ 0 & -\alpha_3 B^* & -\alpha_3 E^* - \mu_2 & 0 \\ 0 & 0 & 0 & -\mu_3 \end{bmatrix}. \tag{9}$$

3.2. Stability Analysis of Equilibria of Model (6)

3.2.1. Immune Checkpoints Equilibrium: $U_0 = \{0, 0, 0, \delta/\mu_3\}$.
The Jacobian matrix $\widehat{J}$ evaluated at U_0 yields

$$\widehat{J}(U_0) = \begin{bmatrix} r & 0 & 0 & 0 \\ 0 & -\mu_1 & 0 & 0 \\ 0 & 0 & -\mu_2 & 0 \\ 0 & 0 & 0 & -\mu_3 \end{bmatrix}. \tag{10}$$

The eigenvalues of $\widehat{J}(U_0)$ are

$$\begin{aligned} \lambda_1 &= r, \\ \lambda_2 &= -\mu_1, \\ \lambda_3 &= -\mu_2, \\ \lambda_4 &= -\mu_3. \end{aligned} \tag{11}$$

Since one of the eigenvalues is always positive, then U_0 is an unstable saddle point. Clinically, U_0 is referred to as the death equilibrium.

3.2.2. BCG-Free Equilibrium: $U_2 = \{\mu_1 r(\delta + k\mu_3)^2/(\alpha_1 a_1 \mu_3{}^2 - r\alpha_2(\delta + k\mu_3)^2), r(\delta + k\mu_3)/\alpha_1\alpha_3, 0, \delta/\mu_3\}$. Assume U_2 exists; that is, $a_1\mu_3 > \alpha_2(\delta + k\mu_3)$; then, substituting U_2 in $\widehat{J}$ yields the following eigenvalues:

$$\begin{aligned} \lambda_1 &= -\mu_3, \\ \lambda_2 &= -\frac{r\alpha_3\delta + \alpha_3 rk\mu_3 + \mu_2\alpha_1\mu_3}{\alpha_{1\mu_3}}, \\ \lambda_3 &= \frac{\sqrt{-(\delta r\mu_3\mu_1 + rk\mu_1\mu_3{}^2)}}{(\delta + k\mu_3)}, \\ \lambda_4 &= \frac{\sqrt{-(\delta r\mu_3\mu_1 + rk\mu_1\mu_3{}^2)}}{(\delta + k\mu_3)}. \end{aligned} \tag{12}$$

Two of the eigenvalues have a real part equal to zero, which signifies neutral stability. Therefore, the equilibrium point U_2 is neutrally stable.

Conclusively, in the absence of treatment, none of the equilibrium points was found to be stable.

3.3. Model without Immune Checkpoints.
Now, we analyze the model without any suppression on the immune system by the immune checkpoints. The model is given by

$$\begin{aligned} \frac{dC}{dt} &= rC - \alpha_1 EC, \\ \frac{dE}{dt} &= a_1 CE + a_2 BE - \alpha_2 EC - \mu_1 E, \\ \frac{dB}{dt} &= b - \alpha_3 EB - \mu_2 B. \end{aligned} \tag{13}$$

The equilibrium points are as follows:

$$\begin{aligned} U_0 &= \left\{0, 0, \frac{b}{\mu_2}\right\}, \\ U_1 &= \left\{0, \frac{ba_2 - \mu_1\mu_2}{\mu_1\alpha_3}, \frac{\mu_1}{a_2}\right\}, \\ U_2 &= \left\{\frac{\mu_1\mu_2\alpha_1 + \alpha_3 r\mu_1 - a_2 b\alpha_1}{(\alpha_3 ra_1 - \alpha_3 r\alpha_2 + \alpha_1\mu_2 a_1 - \alpha_1\alpha_2\mu_2)}, \frac{r}{\alpha_1}, \right. \\ &\quad \left. \frac{b\alpha_1}{(\alpha_3 r + \alpha_1\mu_2)}\right\}. \end{aligned} \tag{14}$$

The equilibrium point U_1 exists only if $ba_2 \geq \mu_1\mu_2$. This means that the cancer cells will disappear if the constant rate of introduction of BCG and activation rate of BCG are bigger than the degradation rates of both the effector cells and the BCG.

The equilibrium point U_2 also exists if

(i) $\mu_2\alpha_1\mu_1 + \alpha_3 r\mu_1 \geq a_2 b\alpha_1$ and $\alpha_3 a_1 r + a_1\alpha_1\mu_2 \geq \alpha_3 ra_2 + \mu_2\alpha_1\alpha_2$,

(ii) $\mu_2\alpha_1\mu_1 + \alpha_3 r\mu_1 \leq a_2 b\alpha_1$ and $\alpha_3 a_1 r + a_1\alpha_1\mu_2 \leq \alpha_3 ra_2 + \mu_2\alpha_1\alpha_2$.

From model (13), we have the following Jacobian matrix:

$$\begin{aligned} &\bar{J}(C^*, E^*, B^*) \\ &= \begin{bmatrix} r - \alpha_1 E^* & -\alpha_1 C^* & 0 \\ a_1 E^* - \alpha_2 E^* & a_2 B^* + a_1 C^* - \alpha_2 C^* - \mu_1 & a_2 E^* \\ 0 & -\alpha_3 B^* & -\alpha_3 E^* - \mu_2 \end{bmatrix}. \end{aligned} \tag{15}$$

3.4. Stability Analysis of Equilibria of Model (13)

3.4.1. BCG Equilibrium: $U_0 = \{0, 0, b/\mu_2\}$.
The eigenvalues of $\bar{J}$ evaluated at U_0 are

$$\begin{aligned} \lambda_1 &= r, \\ \lambda_2 &= \frac{ba_2 - \mu_1\mu_2}{\mu_2}, \\ \lambda_3 &= -\mu_2. \end{aligned} \tag{16}$$

The eigenvalue λ_1 is always positive and the rest are negative. Therefore, the equilibrium point U_0 is an unstable saddle point.

3.4.2. Cancer-Free Equilibrium: $U_1 = \{0, (ba_2 - \mu_1\mu_2)/\mu_1\alpha_3, \mu_1/a_2\}$.
Assume the equilibrium point U_1 exists; then, substituting U_1 in $\bar{J}$ will give the following matrix:

$$\begin{aligned} &\bar{J}(U_1) \\ &= \begin{bmatrix} \dfrac{r\alpha_3\mu_1 - a_1 ba_2 + \mu_2\mu_1}{\alpha_3\mu_1} & 0 & 0 \\ \dfrac{a_1 ba_2 - a_1\mu_2\mu_1 - \alpha_2 ba_2 + \alpha_2\mu_2\mu_1}{\alpha_3\mu_1} & 0 & \dfrac{a_2{}^2 b - a_2\mu_2\mu_1}{\alpha_3\mu_1} \\ 0 & -\dfrac{\alpha_3\mu_1}{a_2} & -\dfrac{ba_2}{\mu_1} \end{bmatrix}. \end{aligned} \tag{17}$$

The eigenvalues of $\bar{J}(U_2)$ are

$$\lambda_1 = \frac{r\alpha_3\mu_1 + \mu_2\mu_1 - a_1 ba_2}{\alpha_3\mu_1},$$

$$\lambda_2 = \frac{-ba_2 + \sqrt{(ba_2)^2 - 4ba_2\mu_1^2 + 4\mu_2\mu_1^3}}{2\mu_1},$$

$$\lambda_3 = \frac{-ba_2 - \sqrt{(ba_2)^2 - 4ba_2\mu_1^2 + 4\mu_2\mu_1^3}}{2\mu_1}.$$

(18)

Now, if

(i) λ_2 and λ_3 are complex roots, then U_1 is a stable fixed point if $a_1 ba_2 > \mu_1(r\alpha_3 + \mu_2)$;

(ii) λ_2 and λ_3 are real roots, then U_1 is a stable fixed point if $ba_2 > \mu_1\mu_2$ and $a_1 ba_2 > \mu_1(r\alpha_3 + \mu_2)$.

But since we already assume that the equilibrium point U_1 exists, then $ba_2 > \mu_1\mu_2$, and we can conclude that U_1 is a stable fixed point if $a_1 ba_2 > \mu_1(r\alpha_3 + \mu_2)$.

This means that the effector cells activated by BCG will eradicate/destroy the cancer cells, if the constant rate of introduction of BCG, recruitment rate of effector cells, and the activation rate of effector cells by BCG are *bigger than or can overcome* the cancer growth rate, the rate of elimination of BCG by effector cells, and the degradation rates of effector cells and BCG altogether. Therefore, to eliminate the cancer, we *increase* the rate of introduction of BCG, rate of recruitment of effector cells, and activation rate of effector cells by BCG and at the same time *decrease* the rate of elimination of BCG by effector cells, degradation rates of both effector cells and BCG, and the cancer growth rate.

3.5. Model with Treatment and Immune Checkpoints. We now consider the dynamics of cancer cells, effector cells BCG, and immune checkpoints (see (5)).

The equilibrium points of model (5) are as follows:

$$U_0 = \left\{ 0, 0, \frac{b}{\mu_2}, \frac{\delta}{\mu_3} \right\},$$

$$U_1 = \left\{ 0, \frac{b\mu_3 a_2 - \mu_2\mu_1\delta - \mu_2\mu_1 k\mu_3}{\mu_1\alpha_3(\delta + k\mu_3)}, \frac{\mu_1(\delta + k\mu_3)}{\mu_3 a_2}, \frac{\delta}{\mu_3} \right\},$$

$$U_2 = \left\{ \frac{\alpha_3 r\delta^2\mu_1 + 2\alpha_3 r\delta\mu_1\mu_3 k + \alpha_3 rk^2\mu_3^2\mu_1 + \mu_2\alpha_1\mu_3^2\mu_1 k + \mu_2\alpha_1\mu_3\mu_1\delta - \mu_3^2 a_2 b\alpha_1}{\alpha_3\mu_3 r\delta a_1 - r\delta^2\alpha_3\alpha_2 - 2\alpha_3 r\delta\alpha_2 k\mu_3 + \alpha_3 rk\mu_3^2 a_1 - \alpha_3 rk^2\mu_3^2\alpha_2 + \mu_2\mu_3^2\alpha_1 a_1 - \mu_2\alpha_1\mu_3\alpha_2\delta - \mu_2\alpha_1\mu_3^2\alpha_2 k}, \right.$$

$$\left. \frac{r(\delta + k\mu_3)}{\mu_3\alpha_1}, \frac{b\alpha_1\mu_3}{\alpha_3 r(\delta + k\mu_3) + \alpha_1\mu_3\mu_2}, \frac{\delta}{\mu_3} \right\}.$$

(19)

The equilibrium point U_1 exists if $\dfrac{b\mu_3 a_2}{\mu_2\mu_1(\delta + k\mu_3)} \geq 1.$ (∗)

Also, U_2 exists if

(i) $\alpha_3 r\delta^2\mu_1 + 2\alpha_3 r\delta\mu_1\mu_3 k + \alpha_3 rk^2\mu_3^2\mu_1 + \mu_2\alpha_1\mu_3^2\mu_1 k + \mu_2\alpha_1\mu_3\mu_1\delta \geq \mu_3^2 a_2 b\alpha_1$ and $\alpha_3\mu_3 r\delta a_1 + \alpha_3 rk\mu_3^2 a_1 + \mu_2\mu_3^2\alpha_1 a_1 \geq r\delta^2\alpha_3\alpha_2 + 2\alpha_3 r\delta\alpha_2 k\mu_3 + \alpha_3 rk^2\mu_3^2\alpha_2 + \mu_2\alpha_1\mu_3\alpha_2\delta + \mu_2\alpha_1\mu_3^2\alpha_2 k$;

(ii) $\alpha_3 r\delta^2\mu_1 + 2\alpha_3 r\delta\mu_1\mu_3 k + \alpha_3 rk^2\mu_3^2\mu_1 + \mu_2\alpha_1\mu_3^2\mu_1 k + \mu_2\alpha_1\mu_3\mu_1\delta \leq \mu_3^2 a_2 b\alpha_1$ and $\alpha_3\mu_3 r\delta a_1 + \alpha_3 rk\mu_3^2 a_1 + \mu_2\mu_3^2\alpha_1 a_1 \leq r\delta^2\alpha_3\alpha_2 + 2\alpha_3 r\delta\alpha_2 k\mu_3 + \alpha_3 rk^2\mu_3^2\alpha_2 + \mu_2\alpha_1\mu_3\alpha_2\delta + \mu_2\alpha_1\mu_3^2\alpha_2 k$.

From model (5), we obtain the following Jacobian matrix:

$$\tilde{J}(C^*, E^*, B^*, P^*) = \begin{bmatrix} r - \dfrac{\alpha_1 E^*}{P^* + k} & \dfrac{\alpha_1 C^*}{P^* + k} & 0 & \dfrac{\alpha_1 E^* C^*}{(P^* + k)^2} \\ \dfrac{a_1 E^*}{P^* + k} - \alpha_2 E^* & \dfrac{a_1 C^* + a_2 B^*}{P^* + k} - \alpha_2 C^* - \mu_1 & \dfrac{a_2 E^*}{P^* + k} & -\dfrac{(a_1 E^* C^* + a_1 B^* C^*)}{(P^* + k)^2} \\ 0 & -\alpha_3 B^* & -\alpha_3 E^* - \mu_2 & 0 \\ 0 & 0 & 0 & -\mu_3 \end{bmatrix}.$$

(20)

3.6. Stability Analysis of Equilibria of Model (5)

3.6.1. BCG and Immune Checkpoints Equilibrium: $U_0 = \{0, 0, b/\mu_2, \delta/\mu_3\}$. The eigenvalues of $\widetilde{J}$ evaluated at U_0 are

$$\lambda_1 = r,$$

$$\lambda_2 = \frac{b\mu_3 a_2 - \mu_2 \mu_1 \delta - \mu_2 \mu_1 \mu_3 k}{\mu_2 (\delta + k\mu_3)},$$

$$\lambda_3 = -\mu_2,$$

$$\lambda_1 = -\mu_3,$$

$$\lambda_2 = \frac{\alpha_3 r \delta^2 \mu_1 + 2\alpha_3 r \delta \mu_3 \mu_1 k + \alpha_3 r k^2 \mu_3{}^2 \mu_1 + \mu_2 \alpha_1 \mu_3{}^2 \mu_1 k + \mu_2 \alpha_1 \mu_3 \mu_1 \delta - \mu_3{}^2 a_2 b \alpha_1}{\alpha_3 \mu_1 (\delta + k\mu_3)^2},$$

$$\lambda_3 = \frac{-b\mu_3 a_2 + \sqrt{(b\mu_3 a_2)^2 + 4\mu_1{}^3 \delta^2 \mu_2 + 8\mu_1{}^3 \delta \mu_2 k\mu_3 + 4\mu_3{}^2 \mu_1{}^3 k^2 \mu_2 - 4\mu_1{}^2 \delta b\mu_3 a_2 - 4\mu_3{}^2 \mu_1{}^2 kba_2}}{2\mu_1 (\delta + \mu_3 k)},$$

$$\lambda_4 = \frac{-b\mu_3 a_2 - \sqrt{(b\mu_3 a_2)^2 + 4\mu_1{}^3 \delta^2 \mu_2 + 8\mu_1{}^3 \delta \mu_2 k\mu_3 + 4\mu_3{}^2 \mu_1{}^3 k^2 \mu_2 - 4\mu_1{}^2 \delta b\mu_3 a_2 - 4\mu_3{}^2 \mu_1{}^2 kba_2}}{2\mu_1 (\delta + \mu_3 k)}.$$

(22)

The equilibrium point U_1 is a stable fixed point if

$$\frac{a_2 b\mu_3}{\mu_1 \mu_2 (\delta + k\mu_3)}$$

$$> \max \left\{ 1, \frac{(r\mu_3 k\alpha_3 + \mu_3 \mu_2 \alpha_1 + \alpha_3 r\delta) \mu_2}{\alpha_1 \mu_3} \right\}.$$

(23)

However, condition $(*)$ is already true; then, U_1 is a stable fixed point if

$$\frac{a_2 b\mu_3}{\mu_1 \mu_2 (\delta + k\mu_3)} > \frac{(r\mu_3 k\alpha_3 + \mu_3 \mu_2 \alpha_1 + \alpha_3 r\delta) \mu_2}{\alpha_1 \mu_3}. \quad (24)$$

3.6.3. Interior Equilibrium: $U_2 = \{C^*, r(\delta + k\mu_3)/\mu_3 \alpha_1, b\alpha_1 \mu_3/(\alpha_3 r(\delta + k\mu_3) + \alpha_1 \mu_3 \mu_2), \delta/\mu_3\}$. The eigenvalues of the Jacobian matrix $\widetilde{J}(U_2)$ are very long, complicated, and difficult to analyze. Therefore, we use numerical simulations to show the stability of the equilibrium point U_2.

4. Numerical Illustrations

In this section, the numerical simulations of the three models will be shown. The aim here is to show the effect of immune checkpoints on the effector cells. We use MATLAB version 2016b to plot the graphs with initial populations of the compartments involved taken to be equal. Other parameters used in the numerical simulations are given in Table 1.

We first plot the graph of model (6) to illustrate what happens in the absence of treatment. As expected, the cancer cells develop with the help of suppression on the effector cells by the immune checkpoints, hence dominating the effector

$$\lambda_4 = -\mu_3.$$

(21)

Since one of the eigenvalues is always positive, then U_0 is an unstable saddle point.

3.6.2. Tumor-Free Equilibrium: $U_1 = \{0, (b\mu_3 a_2 - \mu_2 \mu_1 \delta - \mu_2 \mu_1 k\mu_3)/\mu_1 \alpha_3 (\delta + k\mu_3), \mu_1 (\delta + k\mu_3)/\mu_3 a_2, \delta/\mu_3\}$. Assume this equilibrium point exists; then, the eigenvalues of $\widetilde{J}$ evaluated at U_1 are as follows:

cells and resulting in the growth and maturation of the cancer. Therefore, the numerical simulations of model (6) support this notion as shown in Figure 1.

Next, we show the behavior of model (13) (i.e., without the immune checkpoints). Here, we will see how the effector cells attack and kill the cancer cells as a result of the stimulation/activation by the BCG. Unlike in Figure 1, Figure 2 shows how the growth of the cancer cells is restricted and eventually leads to their extinction by the effector cells.

The general model will now be considered. Despite stimulation and activation of the effector cells by the BCG, the immune suppressors block and deactivate their function; hence, this leads to the reduction of autoimmunity of the effector cells. Therefore, the cancer develops and grows exponentially as shown in Figure 3.

Therefore, comparing Figures 2 and 3, we will notice the effect of immune checkpoints on the effector cells. In Figure 2, the effector cells in the absence of immune suppressors fight the cancer cells, resulting in stopping their development and progression, while Figure 3 shows the progression and development of the cancer cells as a result of the presence of immune suppressors.

5. Conclusion and Discussion

In this paper, we used a system of four nonlinear ordinary differential equations to model the dynamics of cancer cells, effector cells, BCG, and immune checkpoints/suppressors in the immunotherapy of bladder cancer. We derived three possible dynamics from our model. Firstly, the model was analyzed in the absence of treatment and we studied the stability analysis of the equilibria involved. Figure 1 shows

TABLE 1: List of all parameters used in numerical simulations.

| Parameter	Interpretation (units)	Estimated value	Reference
r	Tumor growth rate $t^{-1} = \mathrm{day}^{-1}$	0.0033	Shochat et al., 1999
α_1	Rate of elimination of cancer cells by effector cells cell day^{-1}	1.1×10^{-7}	Kuznetsov et al., 1994
k	Inhibitory parameter	2×10^{3}	Not found
a_1	Recruitment rate of effector cells $t^{-1} = \mathrm{day}^{-1}$	0.25	Sud D. et al., 2006
a_2	Activation rate of effector cells by the BCG cells^{-1} day^{-1}	0.052	Wigginton and Kirschner, 2001
δ	Internal production of immune checkpoints	1.51932×10^{5}	Sandip Banerjee et al., 2015
α_2	Elimination rate of effector cells by cancer cells cells^{-1} day^{-1}	3.45×10^{-10}	Kuznetsov et al., 1994
μ_1	Degradation rate of effector cells $t^{-1} = \mathrm{day}^{-1}$	0.041	Kuznetsov et al., 1994
μ_2	Rate of BCG decay $t^{-1} = \mathrm{day}^{-1}$	0.1	Archuleta et al., 2002
b	Bioeffective concentration of BCG c.f.u./day	6.5×10^{5}	Cheng et al., 2004
α_3	Destruction of BCG by effector cells cells^{-1} day^{-1}	1.25×10^{-7}	Wigginton and Kirschner, 2001
μ_3	Degradation rate of immune checkpoints $t^{-1} = \mathrm{day}^{-1}$	166.32	Sandip Banerjee et al., 2015
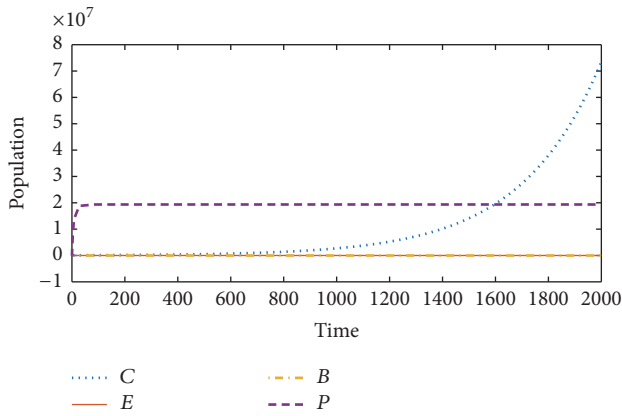
FIGURE 1: Model (6) (without treatment): cancer cells (C) grow exponentially, overcoming the effector cells (E), with the help of immune checkpoints (P).
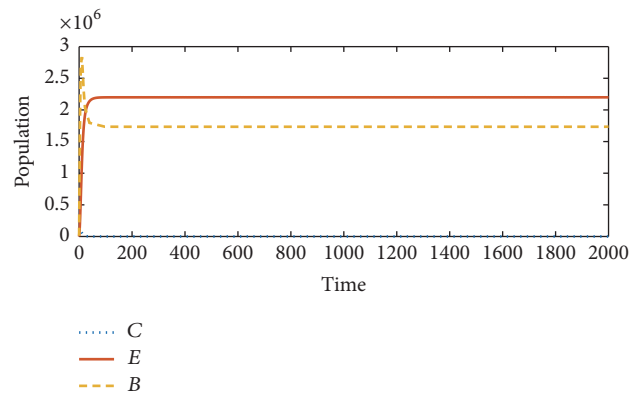

FIGURE 2: Model (13) (without immune suppressors): the effector cells (E) overcome the development of cancer cells (C) as a result of the stimulation and activation by the BCG (B).

how the cancer progressed in the absence of treatment and presence of immune checkpoints/suppressors.

Secondly, we study the model without the immune checkpoints/suppressors. Conditions for stability of the equilibria involved were also given. In the absence of immune checkpoints/suppressors, the activated-effector cells have unlimited freedom to roam about and detect the cancer cells; as a result, they kill them and stop the cancer from progressing. This was shown in Figure 2.

Thirdly, we considered the dynamics of the model with treatment and the immune checkpoints/suppressors.

Conditions for stability of the equilibrium points were given, and Figure 3 shows how the cancer cells grow and develop despite the application of the treatment (BCG). This is believed to be as a result of the blockage and suppression that the effector cells suffered by the immune checkpoints.

Therefore, the figures used in this paper assist in showing the effect of immune checkpoints/suppressors on the effector cells and the treatment at large. To avoid cancer progression and advancement, there is a need for action to block or limit the production of the immune checkpoints. This will take the brakes off the immune system and thereby allow it to mount a stronger and more effective attack against cancer cells.
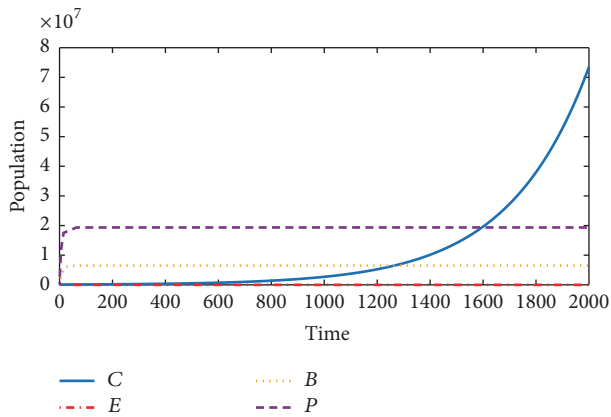
FIGURE 3: Model (5): despite the stimulation of effector cells (E) by the BCG (B), the immune checkpoints (P) block and deactivate the activities of the effector cells, thereby leading to the development and progression of cancer cells (C).

Nivolumab is a drug recently approved by the FDA to be used alone or with other drugs to treat cancer. It is a fully human immunoglobulin (Ig) G4 monoclonal antibody directed against the negative immunoregulatory human cell surface receptor programmed cell death protein-1 (PD-1) with immune checkpoint inhibitory and antineoplastic activities. Nivolumab binds to and blocks the activation or production of immune checkpoints like PD-1. This results in the activation of T-cells and cell-mediated responses against cancer cells. So, the primary role of nivolumab is to block the immune checkpoints from suppressing the immune systems. Hence, this helps in allowing the immune cells to rise against cancer cells without any interference [16, 17].

Conflicts of Interest

The authors declare that they have no conflicts of interest.

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