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ANALYSIS OF IMMUNOTHERAPY MODELS IN THE CONTEXT OF CANCER DYNAMICS

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A basic mathematical model of the immune response when cancer cells are recognized is proposed. The model consists of six ordinary differential equations. It is extended by taking into account two types of immunotherapy: active immunotherapy and adoptive immunotherapy. An analysis of the corresponding models is made to answer the question which of the presented methods of immunotherapy is better. The analysis is completed by numerical simulations which show that the method of adoptive immunotherapy seems better for the patient at least in some cases.

Keywords: dynamic systems, immune system, cancer, vaccination

1. Introduction

The treatment of cancer is one of the most challenging problems of modern medicine. An ideal treatment method should fulfil two basic conditions. First, it should destroy cancer cells in the entire body. Second, it should distinguish between cancerous and healthy cells. Immunotherapy seems to be the method that best fulfils both of these requirements (Chen and Wu, 1998).

The immune system comprises many types of lymphocytes which effectively destroy foreign strange cells after activation. Some lymphocytes even exhibit natural cytotoxicity, i.e., they do not require activation, e.g., Natural Killer (NK) cells. B and T lymphocytes have a wide range of antigen receptors, which allows the immune system to identify foreign antigens and to distinguish cancer cells.

The stimulation of an immune system in order to provide an effective treatment of cancer diseases can by realized with the aid of vaccinations. It is necessary to clarify that the term "vaccine" with reference to cancer therapy does not mean the prevention of the disease as is commonly understood. In the case of the application of vaccine against the cancer we still have to deal with the activation of the immune system. The difference is in the fact that the vaccine now has the character of treatment, not that of prevention.

In this paper we propose a mathematical model of the immune system response to the identification of cancer antigens. Some other models can be found in (Foryś, 2002; Villasana, 2001). The presented model confirms the well-known fact that if the cancer is very aggressive, then despite the identification of cancer cells, the immune system is unable to stop the development of the neoplasm. In this case cancer vaccine can be very useful. The proposed model is extended to allow the comparison of various types of anti-tumor vaccine. A mathematical analysis of the proposed models is carried out together with a numerical simulation to determine which of the treatment methods is more effective. In these models the three-dimensional space structure of the neoplasm is not taken into account. Our models can be used for cancers characterized by easy access to cells, and best to disseminated cancer cells. We mention, however, that in the literature one can find spatially structured models (Greenspan, 1972).

2. Basic Model

The model describes the dynamics of cancer cells and some selected elements of the immune system whose role seems significant, i.e., NK, LAK, T helper cells, B lymphocytes and cytotoxic T lymphocytes (CTL). The components are identified by their concentration without taking into account their three-dimensional distribution. We denote by $T(t)$, $K(t)$, $L(t)$, $H(t)$, $C(t)$ and $B(t)$ the numbers of cancer cells, NK and LAK lymphocytes, helper and cytotoxic T cells and B cells at time $(t > 0)$, respectively.

The total number of cancer cells in a body depends on the rates at which they divide and are destroyed by the immune system. We assume that the number of cancer cells increases exponentially by division and that their decline occurs mainly through the action of NK and LAK cells. We can describe these processes by (2). The term a_1T represents the growth in the number of cancer cells due to proliferation, where a_1 is the tumor's proliferation rate. The next two terms $\alpha_1 TK$ and $\beta_1 TL$ describe the reduction of tumor cells by the activity of NK and LAK lymphocytes (the direct destruction of cancer cells and their elimination through the production of the tumor necrosis factor (TNF)). It is assumed that the amount of the TNF produced by NK and LAK cells is proportional to their number, but LAK cells are more effective in the destruction of cancer cells (Jakóbisiak, 1995). Therefore we have

$$\alpha_1 < \beta_1. \tag{1}$$

The model also takes into account the activity of B lymphocytes and cytotoxic T lymphocytes. The first factor affects the tumor cells mainly by the mediation of the production of antibodies. Due to the contact with antigens, B cells are activated and begin producing antibodies. This process is described by the term $\gamma_1 TB$. The last term in (2), i.e., $\delta_1 TC$ is responsible for the loss of tumor cells due to the activity of T cytotoxic cells. Finally, we have the following equation:

$$\dot{T} = a_1T - \alpha_1 TK - \beta_1 TL - \gamma_1 TB - \delta_1 TC. \tag{2}$$

NK cells are derived from precursor cells that mature in the bone marrow. A certain number of mature lymphocytes circulate in the bloodstream, and some of them enter lymphatic organs and tissues. Therefore it can be said that the number of these lymphocytes depends on the rate at which they enter the bloodstream. We can describe it using the quantity s_2. The presence of foreign antigens in a tissue may cause an additional increase in the number of NK cells, mainly as a result of the intensified recruitment of primary cells. This effect can be modeled by the term $\beta_2 T$. The number of NK cells decreases mainly through apoptosis. We describe this by the term $-dK$, where d is the mortality coefficient. Some NK cells may undergo activation and transformation into LAK cells. This process occurs in the presence of large amounts of interleukin II, which is one of the cytokines produced by T helper cells to activate other immune cells. We assume that the amount of interleukin II is proportional to the number of T helper cells producing it. This phenomenon is described by the term $-\alpha KH$. The coefficient α is relatively small because the amount of interleukin II needed to transform NK lymphocytes into LAK cells is very high. Finally, we obtain the following equation:

$$\dot{K} = s_2 - dK - \alpha KH + \beta_2 T. \tag{3}$$

We assume that the increase in the number of LAK cells occurs as a result of the transformation of NK cells activated by the high level of interleukin II. This process is described by the term $+\alpha KH$. The decrease in the number of these cells is a result of apoptosis. This effect is represented by the term $-dL$. The equation describing LAK cells dynamic reads as follows:

$$\dot{L} = -dL + \alpha KH. \tag{4}$$

Similarly to cytotoxic T lymphocytes and B lymphocytes, T helper cells arise in the bone marrow, and then proceed through the process of maturation to enter finally the circulatory system (Jakóbisiak, 1995). Its main goal is to stimulate the immune response. The dynamics of this population are described by the equation

$$\dot{H} = s_4 - d_4 H + \alpha_4 H(t-\tau)\big[T(t-\tau) - q_4 T^2(t-\tau)\big]. \tag{5}$$

The number of T helper cells depends mainly on the rate at which new lymphocytes are produced in the bone marrow and the rate of apoptosis. These phenomena are described respectively by the terms s_4 and $-d_4H$. The parameter d_4 is the mortality coefficient. The next term, i.e., $\alpha_4 H(t-\tau)[T(t-\tau) - q_4 T^2(t-\tau)]$ describes the phenomenon of the increasing number of tumor cells due to the stimulation by the presence of identified tumor antigens. In our model we assume that in patients with advanced, metastatic cancer, the strength of the immune reaction is significantly reduced (Jakóbisiak, 1995). This is the reason behind the occurence of the logistic component in the considered equation. The term $H(t-\tau)$ represents the relationship between the proliferation rate of T helper cells and their number. This relationship is twofold. First, the proliferation rate depends on the number of cells which can proliferate. Next, the proliferation process needs the presence of some interleukins which are mainly produced by some subpopulation of T helper cells, the so called Th1. In the equations describing the dynamics of T helper cells, cytotoxic T lymphocytes, and B lymphocytes, the time $\tau > 0$ is taken into consideration for the successive divisions of lymphocytes, because these processes are relatively long.

The number of B lymphocytes depends on the level of their production in the bone marrow, the rate at which they are activated by an antigen and at which they proliferate, and their characteristic level of apoptosis. We describe it using the following equation:

$$\begin{aligned}\dot{B} = s_5 - d_5 B + \alpha_5 H(t-\tau)\big[T(t-\tau)\\ - q_5 T^2(t-\tau)\big]B(t-\tau).\end{aligned} \tag{6}$$

Similarly to (3) and (5), the constant flow of B cells is modeled by the term s_5. The expression $-d_5B$ is responsible for the loss of B cells due to apoptosis. In the next term, i.e. $\alpha_5 H(t-\tau)[T(t-\tau) - q_5 T^2(t-\tau)]B(t-\tau)$, we describe the increase in the number of B cells caused by activation. As has previously been mentioned, T helper cells play a very important role in the regulation of the immunological response. They facilitate activation, proliferation and the differentiation of B helper cells, as well as the precursors of T cytotoxic cells. Therefore the term $H(t-\tau)$ occurs in the expression considered. Moreover, the proliferation rate depends on the number of cells which can divide. This process could be represented by the term $B(t-\tau)$. The logistic term is used once again for the same reason as in the previous case. We assume that the time needed for cells to divide is comparable. Therefore we use the same time delay τ.

The equation describing the dynamics of cytotoxic T lymphocytes is analogous to the previous one. The activity mechanisms of B and T lymphocytes are completely different, but the appearance, the loss through apoptosis, and the activation process can be modeled in the same way. Thus we obtain the following equation:

$$\begin{aligned}\dot{C} &= s_6 - d_6 C + \alpha_6 H(t-\tau)\big[T(t-\tau) \\ &\quad - q_6 T^2(t-\tau)\big] C(t-\tau). \end{aligned} \tag{7}$$

Finally, we study the system of six ordinary differential equations with time delay that consists of Eqns. (2)–(7). We assume that all coefficients are positive, but in Section 5, in order to simplify the model, we set $q_4 = 0$.

3. Extension of the Model to Include Immunotherapy

Of great interest is to study the possibility of making use of the immune system in tumor therapy. This raises the activity of the tumor necrosis factor, TNF. It affects the tumor cells, induces alterations in the tumor's blood vessels and stimulates the immune response of other lymphocytes. While operating directly on tumor cells, the TNF causes their decomposition, stops their proliferation and induces differentiation. In cells subjected to the activity of the TNF we can observe DNA spallation and death similar to apoptosis.

Both of the examined ways of the activation of the immune system are aimed at stimulating it mainly to produce the TNF. The first way is based on introducing T helper cells into the patient's body. These cells secrete interleukin causing the transformation of NK into LAK cells. The second way is the administration of vaccine that stimulates the production of T helper cells.

The first method constitutes a variant of a wider approach called adoptive immunotherapy. The patient is given immune cells that have been activated outside his body. In this case T helper cells can identify cancer antigens. For simplicity, we assume that in both cases the vaccine is introduced into the body in a continuous way. The constant influx of T helper lymphocytes, caused by the vaccination, is reflected by the term v_4 and added into the equation describing T helper cell dynamics. Finally, the model with the adoptive immunotherapy is the following:

$$\begin{aligned}\dot{T} &= a_1 T - \alpha_1 TK - \beta_1 TL - \gamma_1 TB - \delta_1 TC,\\ \dot{K} &= s_2 - dK - \alpha KH + \beta_2 T,\\ \dot{L} &= -dL + \alpha KH,\\ \dot{H} &= s_4 - d_4 H + \alpha_4 H(t-\tau)\big[T(t-\tau)\\ &\quad - q_4 T^2(t-\tau)\big] + v_4,\\ \dot{B} &= s_5 - d_5 B + \alpha_5 H(t-\tau)\big[T(t-\tau)\\ &\quad - q_5 T^2(t-\tau)\big] B(t-\tau),\\ \dot{C} &= s_6 - d_6 C + \alpha_6 H(t-\tau)\big[T(t-\tau)\\ &\quad - q_6 T^2(t-\tau)\big] C(t-\tau). \end{aligned} \tag{8}$$

The administrated lymphocytes are an additional source of lymphocytes needed to activate other components of the immune system and compounds from the group of interleukins, especially interleukin II.

The second method is an example of active immunotherapy. The patient is given vaccine, usually in the form of attenuated cancer cells or their antigens. The administration of the vaccine activates T helper cells. We describe it by the following equation:

$$\dot{V} = s_7 - d_7 V. \tag{9}$$

The rate at which the vaccine enters the bloodstream can be described using the term s_7. The second expression, $-d_7 V$, is responsible for the loss of vaccine. In the presence of the vaccine, we have to change the equation describing the dynamics of T helper cells. The additional term $\beta_4 H(t-\tau)V(t-\tau)$ describes the increase in the proliferation of T helper cells as a result of the administrated vaccine. Finally, we obtain the following model:

$$\begin{aligned}\dot{T} &= a_1 T - \alpha_1 TK - \beta_1 TL - \gamma_1 TB - \delta_1 TC,\\ \dot{K} &= s_2 - dK - \alpha KH + \beta_2 T,\\ \dot{L} &= -dL + \alpha KH,\\ \dot{H} &= s_4 - d_4 H + \alpha_4 H(t-\tau)\big[T(t-\tau)\\ &\quad - q_4 T^2(t-\tau)\big] + \beta_4 H(t-\tau)V(t-\tau),\\ \dot{B} &= s_5 - d_5 B + \alpha_5 H(t-\tau)\big[T(t-\tau)\\ &\quad - q_5 T^2(t-\tau)\big] B(t-\tau),\\ \dot{C} &= s_6 - d_6 C + \alpha_6 H(t-\tau)\big[T(t-\tau)\\ &\quad - q_6 T^2(t-\tau)\big] C(t-\tau),\\ \dot{V} &= s_7 - d_7 V. \end{aligned} \tag{10}$$

4. Simplified Models

Equations (8) and (10) are quite complicated. In order to analyze the behavior of their solutions, we simplified them by excluding certain components of the immune response. We assume that both the antibodies and the cytokines produced by cytotoxic T lymphocytes act only minimally on the populations of cancer cells. Therefore the expressions and equations describing these lymphocytes are excluded from both models.

The next simplification is the assumption that $\tau = 0$. It is justified by the fact that in a situation where a menace is recognized, the specific immune response is very quick. Therefore we neglect the cell division time in the models considered.

Consider the model with adoptive immunotherapy. Instead of constants s_4 and v_4, we introduce the new constant s_4^* into Eqn. (5). This constant is responsible for the increase in the number of T helper cells due to the rate at which these lymphocytes enter the bloodstream and vaccination. After these simplifications the model is as follows:

$$\begin{aligned}\dot{T} &= a_1 T - \alpha_1 TK - \beta_1 TL,\\ \dot{K} &= s_2 - dK - \alpha KH + \beta_2 T,\\ \dot{L} &= -dL + \alpha KH,\\ \dot{H} &= s_4^* - d_4 H + \alpha_4 H[T - q_4 T^2].\end{aligned} \tag{11}$$

Consider now the model with active immunotherapy. Equation (9), describing the dynamics of the administrated vaccine, is independent of other equations. Solving it, we obtain

$$V(t) = \frac{s_7}{d_7}(1 - e^{-d_7 t}), \quad V(0) = 0, \tag{12}$$

which exponentially tends to the value s_7/d_7. Therefore, still making a simplification, we assume that the amount of vaccine does not change in time and is equal to s_7/d_7. Hence we find that the term $\beta_4 V$ can be approximated by a constant value s_7/d_7.

For this reason, instead of the term $d_4 H$, we can consider the expression $d_4^* H$. The new coefficient d_4^* reflects the loss of T helper cells caused by the apoptosis as well as the increase of this lymphocytes due to the administrated vaccine. We would like to stress that the coefficient d_4^* is the only parameter in both models which can take a negative value. Finally, we obtain the following model:

$$\begin{aligned}\dot{T} &= a_1 T - \alpha_1 TK - \beta_1 TL,\\ \dot{K} &= s_2 - dK - \alpha KH + \beta_2 T,\\ \dot{L} &= -dL + \alpha KH,\\ \dot{H} &= s_4 - d_4^* H + \alpha_4 H[T - q_4 T^2].\end{aligned} \tag{13}$$

We emphasize once more the differences between these two models because formally they are the same. In the case of the model with adoptive immunotherapy we have $s_4^* = s_4 + v_4$, where $v_4 > 0$, while in the case of the model with active immunotherapy $d_4^* = d_4 - s_7/d_7$, where $s_7/d_7 > 0$. We want to study the behavior of the solutions to the models when the coefficients v_4, s_7 and d_7 change.

In Section 6 we perform numerical simulations for Eqns. (11) and (13). However, to make a mathematical analysis, we assume that $q_4 = 0$. Therefore, in Section 5 we consider the model with adoptive immunotherapy

$$\begin{aligned}\dot{T} &= a_1 T - \alpha_1 TK - \beta_1 TL,\\ \dot{K} &= s_2 - dK - \alpha KH + \beta_2 T,\\ \dot{L} &= -dL + \alpha KH,\\ \dot{H} &= s_4^* - d_4 H + \alpha_4 HT.\end{aligned} \tag{14}$$

and the model with active immunotherapy

$$\begin{aligned}\dot{T} &= a_1 T - \alpha_1 TK - \beta_1 TL,\\ \dot{K} &= s_2 - dK - \alpha KH + \beta_2 T,\\ \dot{L} &= -dL + \alpha KH,\\ \dot{H} &= s_4 - d_4^* H + \alpha_4 HT.\end{aligned} \tag{15}$$

5. Analysis of the Simplified Models

First, we study the existence, uniqueness and nonnegativity of solutions. We should stress that only nonnegative solutions (corresponding to nonnegative initial data) make biological sense.

In order to prove the uniqueness of solutions, we use the Picard-Lindelöf theorem. To investigate the stability of the solutions, we use the method of linearizations (Hartman, 1964).

Proposition 1. (Global existence and uniqueness.) *If the initial values T_0, K_0, L_0 and H_0 are nonnegative, then there exist nonnegative, unique global solutions to Eqns. (14) and (15).*

Proof. We can outline the proof only for Eqn. (14), noting that the corresponding proof for Eqn. (15) is the same. The existence and uniqueness of solutions follows directly from the Picard-Lindelöf theorem. Now we should prove that the solutions are nonnegative for the nonnegative initial data.

Assume that $(T(t), K(t), L(t), H(t))$ is a solution to (14). Then $T(t)$ satisfies the following integral equation:

$$T(t) = T_0 e^{\int_0^t \left(a_1 - \alpha_1 K(s) - \beta_1 L(s)\right)\, \mathrm{d}s}. \tag{16}$$

Since $T_0 \geq 0$, we have $T(t) \geq 0$ for $t \geq 0$ for which the solution exists.

From
$$\dot{K} \geq -dK - \alpha KH, \tag{17}$$
we obtain
$$K(t) \geq K_0 e^{\int_0^t \left(-d-\alpha H(s)\right)\,\mathrm{d}s}. \tag{18}$$

If $K_0 \geq 0$, then we have $K(t) \geq 0$ for any $t \geq 0$ for which the solution exists.

Since
$$\dot{H} \geq H(-d_4 + \alpha_4 T) \tag{19}$$
and
$$H(t) \geq H_0 e^{\int_0^t \left(-d_4+\alpha_4 T(s)\right)\,\mathrm{d}s}, \tag{20}$$
if the initial value H_0 is nonnegative, then so is $H(t)$.

From $\alpha KH \geq 0$ we have
$$L(t) \geq L_0 e^{\int_0^t (-d)\,\mathrm{d}s} = L_0 e^{-\mathrm{d}t}, \tag{21}$$
and $L(t) \geq 0$ for $L_0 \geq 0$ and $t \geq 0$ for which the solution exists. Therefore the solution $(T(t), K(t), L(t), H(t))$ is nonnegative for nonnegative initial data.

Consider the first equation of (14). It follows that
$$T(t) \leq T_0 e^{a_1 t}. \tag{22}$$

The right-hand side of (22) is bounded on any $[0,t)$. Hence $T(t)$ is also bounded on $[0,t)$.

Now consider the inequality
$$\dot{K} \leq s_2 - dK + \beta_2 T. \tag{23}$$

It implies
$$K(t) \leq e^{-\mathrm{d}t}\Big(K_0 + \int_0^t e^{\mathrm{d}s}\big[s_2 + \beta_2 T(s)\big]\,\mathrm{d}s\Big). \tag{24}$$

Using (22), we obtain
$$K(t) \quad \leq \quad K_0 e^{-dt} + \tfrac{s_2}{d} + T_0 \tfrac{\beta_2}{d} e^{a_1 t}. \tag{25}$$

Thus $K(t)$ is bounded on any compact time interval.

We have
$$\dot{H} \leq s_4^* + \alpha_4 HT, \tag{26}$$
and
$$H(t) \leq \Big(H_0 + s_4^* \int_0^t e^{-\alpha_4 \int_0^\tau T(s)\,\mathrm{d}s}\mathrm{d}\tau\Big) e^{\alpha_4 \int_0^t T(s)\,\mathrm{d}s}. \tag{27}$$

We use again (22) and obtain
$$H(t) \leq (H_0 + s_4^* t) e^{\frac{T_0 \alpha_4}{a_1} e^{a_1 t}}. \tag{28}$$

Thus $H(t)$ is bounded on any $[0,t)$.

The third equation of (14) can be estimated in the following way:
$$\dot{L} \leq dL + \alpha KH. \tag{29}$$

Hence
$$L(t) \leq e^{\mathrm{d}t}\Big(L_0 + \alpha \int_0^t e^{-\mathrm{d}s} K(s)H(s)\,\mathrm{d}s\Big). \tag{30}$$

$H(t)$ and $K(t)$ are nonnegative and therefore (30) implies that $L(t)$ is bounded on every $[0,t)$. Thus the solution $(T(t), K(t), L(t), H(t))$ is global in time. ■

Equation (14) has three stationary points which are
$$A_0 = \Big[0, \frac{s_2 d_4}{dd_4 + \alpha s_4^*}, \frac{\alpha s_2 s_4^*}{d(dd_4 + \alpha s_4^*)}, \frac{s_4^*}{d_4}\Big], \tag{31}$$
$$A_1 = \Big[pK_1 + q, K_1, \frac{\alpha r}{d}(a_1 - \alpha_1 K_1), \frac{r(a_1 - \alpha K_1)}{K_1}\Big], \tag{32}$$
$$A_2 = \Big[pK_2 + q, K_2, \frac{\alpha r}{d}(a_1 - \alpha_1 K_2), \frac{r(a_1 - \alpha K_2)}{K_2}\Big], \tag{33}$$
where
$$K_1 = \frac{a_1 u + \alpha_1 w + s_4^* + \sqrt{\Delta}}{2\alpha_1 u},$$
$$K_2 = \frac{a_1 u + \alpha_1 w + s_4^* - \sqrt{\Delta}}{2\alpha_1 u}, \tag{34}$$
$$\Delta = (-a_1 u + \alpha_1 w + s_4^*)^2 + 4 a_1 s_4^* u, \tag{35}$$
$$u = \alpha_4 pr, \quad w = r(d_4 - \alpha_4 q),$$
$$p = \frac{d(\beta_1 - \alpha_1)}{\beta_1 \beta_2}, \quad q = \frac{a_1 d - s_2 \beta_1}{\beta_1 \beta_2}, r = \frac{d}{\alpha \beta_1}. \tag{36}$$

Note that the new parameters u, p and r are always positive (cf. (1)). It is also easily seen that all coordinates of the point A_0 are nonnegative. It remains to check the nonnegativity of the coordinates of the points A_1 and A_2.

Proposition 2. *The coordinates of the point A_1 do not satisfy the nonnegativity condition.*

Proof. Consider the third coordinate of A_1.[1] L_1 is nonnegative if and only if $a_1 - \alpha_1 K_1 \geq 0$, which leads to
$$a_1 u - \alpha_1 w - s_4^* \geq \sqrt{\Delta}. \tag{37}$$

[1] The consecutive coordinates of the point A_1 are denoted by T_1, K_1, L_1 and H_1, and those of the point A_2 by T_2, K_2, L_2 and H_2.

Note that (37) is satisfied only if $a_1u - \alpha_1 w - s_4^* > 0$. Assuming this, we obtain the condition $a_1 u s_4^* \le 0$, which is never satisfied. Therefore, the point A_1 does not satisfy the basic condition of the nonnegativity of the coordinates. ∎

Proposition 3. *The coordinates of the point A_2 are nonnegative if and only if*

$$-\frac{a_1 d_4 pr}{d_4\alpha_1 r + s_4^*} \le q < \frac{d_4}{\alpha_4}. \tag{38}$$

Proof. Proceeding similarly to the case of L_1, we obtain that the coordinate L_2 is positive. The coordinate K_2 is nonnegative if and only if

$$a_1 u + \alpha_1 w + s_4^* \ge \sqrt{\Delta}. \tag{39}$$

This yields $a_1u + \alpha_1 w + s_4^* > 0$. Squaring both sides of (39), we obtain $a_1\alpha_1 wu \ge 0$. This inequality is satisfied if and only if $w \ge 0$, i.e., $q \le d_4/\alpha_4$. If $w \ge 0$, then $a_1u + \alpha_1 w + s_4^* > 0$. Hence K_2 is nonnegative if and only if $q \le d_4/\alpha_4$.

H_2 is nonnegative if $a_1 - \alpha_1 K_2 \ge 0$ and $K_2 > 0$, or if $a_1 - \alpha_1 K_2 \le 0$ and $K_2 < 0$. The second case can be rejected. The inequality $a_1 - \alpha_1 K_2 \ge 0$ is satisfied. Therefore $H_2 \ge 0$ if and only if $K_2 > 0$, i.e., $q < d_4/\alpha_4$.

T_2 is nonnegative if and only if $pK_2 + q \ge 0$. This is certainly true when $q \ge 0$. Assume that $q < 0$. The inequality $pK_2 + q \ge 0$ leads to

$$a_1u + \alpha_1 w + s_4^* + 2\alpha_1\alpha_4 qr \ge \sqrt{\Delta}, \tag{40}$$

which is fulfilled only if $a_1u + \alpha_1 w + s_4^* + 2\alpha_1\alpha_4 qr > 0$. Replacing w by $r(d_4 - \alpha_4 q)$ and performing some calculations, we obtain

$$q > -\Big(\frac{a_1\alpha_4 pr + d_4\alpha_1 r + s_4^*}{\alpha_1\alpha_4 r}\Big). \tag{41}$$

This implies $a_1\alpha_4 prq + \alpha_1 qr(d_4 - \alpha_4 q) + s_4^* q + \alpha_1\alpha_4 q^2 r \ge -a_1 d_4 pr + a_1\alpha_4 pqr$. Hence

$$q \ge -\frac{a_1 d_4 pr}{d_4\alpha_1 r + s_4^*}. \tag{42}$$

Thus we obtain

$$\frac{d_4}{\alpha_4} > q, \quad q > -\frac{a_1\alpha_4 pr + d_4^*\alpha_1 r + s_4}{\alpha_1\alpha_4 r},$$

$$\text{and} \quad q \ge -\frac{a_1 d_4^* pr}{d_4^*\alpha_1 r + s_4}. \tag{43}$$

Clearly, in this case

$$-\frac{a_1 d_4 pr}{d_4\alpha_1 r + s_4^*} > -\frac{a_1\alpha_4 pr + d_4^*\alpha_1 r + s_4}{\alpha_1\alpha_4 r}.$$

Hence

$$-\frac{a_1 d_4 pr}{d_4\alpha_1 r + s_4^*} \le q < \frac{d_4}{\alpha_4}. \tag{44}$$

∎

Proposition 4. *If*

$$s_4^* > \frac{dd_4(s_2\alpha_1 - a_1 d)}{\alpha(a_1 d - s_2\beta_1)}, \tag{45}$$

then A_0 is a stable node. If

$$s_4^* < \frac{dd_4(s_2\alpha_1 - a_1 d)}{\alpha(a_1 d - s_2\beta_1)}, \tag{46}$$

then A_0 is a saddle.

Proof. The Jacobian for the point A_0 is as follows:

$$\begin{pmatrix} a_1 - \alpha_1\frac{s_2 d_4}{dd_4+\alpha s_4^*} - \beta_1\frac{\alpha s_2 s_4^*}{d(dd_4+\alpha s_4^*)} & 0 & 0 & 0 \\ \beta_2 & -\alpha\frac{s_4^*}{d_4} - d & 0 & -\alpha\frac{s_2 d_4}{dd_4+\alpha s_4^*} \\ 0 & \alpha\frac{s_4^*}{d_4} & -d & \alpha\frac{s_2 d_4}{dd_4+\alpha s_4^*} \\ \alpha_4\frac{s_4^*}{d_4} & 0 & 0 & -d_4 \end{pmatrix}. \tag{47}$$

Hence the characteristic polynomial is

$$W_0(\lambda) = \Big(a_1 - \frac{\alpha_1 s_2 d_4}{dd_4+\alpha s_4^*} - \frac{\beta_1\alpha s_2 s_4^*}{d(dd_4+\alpha s_4^*)} - \lambda\Big) \times\Big(-\frac{\alpha s_4^*}{d_4} - d - \lambda\Big)(-d-\lambda)(-d_4-\lambda). \tag{48}$$

$W_0(\lambda)$ has only real roots:

$$\lambda_1 = a_1 - \alpha_1\frac{s_2 d_4}{dd_4+\alpha s_4^*} - \beta_1\frac{\alpha s_2 s_4^*}{d(dd_4+\alpha s_4^*)},$$

$$\lambda_2 = -\alpha\frac{s_4^*}{d_4} - d, \quad \lambda_3 = -d, \quad \lambda_4 = -d_4.$$

It is obvious that $\lambda_2, \lambda_3, \lambda_4 < 0$. If $\lambda_1 < 0$, then the point A_0 is a stable node. If $\lambda_1 > 0$, then A_0 is a saddle.

The sign of λ_1 depends on the magnitude of s_4^*. Consider the auxiliary function

$$f(s_4^*) = a_1 - \alpha_1\frac{s_2 d_4}{dd_4+\alpha s_4^*} - \beta_1\frac{\alpha s_2 s_4^*}{d(dd_4+\alpha s_4^*)}. \tag{49}$$

The derivative of this function is always negative (because $\alpha_1 < \beta_1$). Thus the function $f(s_4^*)$ is strictly decreasing, and it is equal to zero if

$$s_4^* = \frac{dd_4(s_2\alpha_1 - a_1 d)}{\alpha(a_1 d - s_2\beta_1)}.$$

∎

The case when A_0 is a stable node is good for the patient. It is equivalent to a total recovery. When A_0 is a saddle, then the values of K, L and H stabilize in time, but the number of cancer cells increases. It is very harmful for the patient. If the administrated vaccine v_4 is greater than $\frac{dd_4(s_2\alpha_1 - a_1 d)}{\alpha(a_1 d - s_2\beta_1)} - s_4$, then the patient has a chance for a total recovery.

The task is now to examine the stability of the point A_2. The Jacobi matrix at this point is

$$\begin{pmatrix} -\lambda & -\alpha_1 T_2 & -\beta_1 T_2 & 0 \\ \beta_2 & -\alpha H_2 - d - \lambda & 0 & -\alpha K_2 \\ 0 & \alpha H_2 & -d-\lambda & \alpha K_2 \\ \alpha_4 H_2 & 0 & 0 & -d_4 + \alpha_4 T_2 - \lambda \end{pmatrix}. \quad (50)$$

The characteristic polynomial has the form

$$\begin{aligned} W_2(\lambda) = \lambda^4 + \lambda^3 \left(\frac{s_4^*}{H_2} + 2d + \alpha H_2 \right) \\ + \lambda^2 \left(2d \frac{s_4^*}{H_2} + \alpha s_4^* + d^2 + \alpha d H_2 + \alpha_1 \beta_2 T_2 \right) \\ + \lambda \Bigg(\alpha_1 \beta_2 d T_2 + \alpha \beta_1 \beta_2 H_2 T_2 + \alpha_1 \beta_2 s_4^* \frac{T_2}{H_2} \\ + d\alpha s_4^* T_2 + d^2 \frac{s_4^*}{H_2} + \alpha \alpha_4 K_2 H_2 T_2 (\beta_1 - \alpha_1) \Bigg) \\ + T_2 \Bigg(d\alpha_1 \beta_2 s_4^* \frac{T_2}{H_2} + \alpha \beta_1 \beta_2 s_4^* T_2 \\ + \alpha \alpha_4 K_2 H_2 (\beta_1 - \alpha_1) \Bigg). \end{aligned} \quad (51)$$

Our goal is to determine the signs of its roots. We start with the observation that all the coefficients of this polynomial are positive.

Using the Fourier theorem (Turowicz, 1967), we obtain the following result:

Corollary 1. *The characteristic polynomial (51) has no nonnegative real roots. Moreover, it has exactly zero, two or four negative, real roots.*

Assume that $W_2(\lambda)$ has complex roots. For simplicity, denote by c_3, c_2, c_1 and c_0 the subsequent coefficients of (51). The polynomial $W_2(\lambda)$ can be represented as a product of two polynomials of the second degree. We have

$$W(\lambda) = (\lambda^2 + m_1\lambda + n_1)(\lambda^2 + m_2\lambda + n_2), \quad (52)$$

where $c_3 = m_1 + m_2$, $c_2 = m_1 m_2 + n_1 + n_2$, $c_1 = n_1 m_2 + n_2 m_1$ and $c_0 = n_1 n_2$.

It is easy to see that if m_1, m_2, n_1 and n_2 are positive, then all the roots of W_2 have negative real parts. But the situation when m_1 or m_2 are negative is also possible. We assume that $m_2 < 0$. If the absolute value of m_2 is relatively small such that the following inequalities hold:

$$\begin{aligned} c_3 &= m_1 + m_2 > 0, \\ c_2 &= m_1 m_2 + n_1 + n_2 > 0, \\ c_1 &= n_1 m_2 + n_2 m_1 > 0, \end{aligned} \quad (53)$$

then the solution is unstable.

Corollary 2. *The characteristic polynomial (51) always has at least two roots which have negative real parts. The point A_2 is then stable or it is a saddle.*

From a medical point of view, the stability of this point means that the disease does not progress, but the patient is not definitely cured. The instability of this point means that the immune system is unable to respond adequately. The patient cannot reach the status of homeostasis (Kuby, 1997).

We now turn to Eqns. (15). It is obvious that the stationary points are the same as for Eqns. (14).[2] But now the parameter d_4^* can have a negative value, so the nonnegativity conditions for this point may have to be changed.

A necessary and sufficient condition for the nonnegativity of all the coordinates of the point A_0 is $d_4^* > 0$. If $d_4^* = 0$, then the stationary point A_0 does not exist. The number of cancer cells, NK and LAK lymphocytes stabilize, but the number of T helper cells increases and finally reaches a size which can be dangerous for the patient, because T helper cells produce cytokinin which can be toxic for the patient in large amounts (Kuby, 1997).

Proposition 5. *The coordinates of the point A_1 do not satisfy the nonnegativity condition. The coordinates of the point A_2 are nonnegative if and only if one of the following systems of conditions is satisfied:*

$$-\frac{a_1 d_4 pr}{d_4 \alpha_1 r + s_4^*} \le q < \frac{d_4}{\alpha_4} \quad \textit{and} \quad 0 < d_4^*, \quad (54)$$

$$-\frac{a_1 \alpha_4 pr + d_4^* \alpha_1 r + s_4}{\alpha_1 \alpha_4 r} < q < \frac{d_4}{\alpha_4}$$

$$\textit{and} \quad -\frac{a_1 \alpha_4 pr + s_4}{2\alpha_1 r} < d_4^* < -\frac{s_4}{\alpha_1 r}. \quad (55)$$

Proof. If $d_4^* \ge 0$, then the nonnegativity conditions of A_1 and A_2 are the same as in (14). Assuming $d_4^* < 0$, we have $L_1 < 0$ and $L_2 > 0$. Hence we can exclude the

[2] Instead of s_4^* and d_4 we have s_4 and d_4^*, respectively.

point A_1. The coordinates K_2 and H_2 are nonnegative if $w > 0$, i.e., $q < d_4/\alpha_4$. The task is now to find conditions for the nonnegativity of T_2. From the analysis of (14) we have that (43) has to be fulfilled. Clearly, it is necessary that

$$\frac{d_4^*}{\alpha_4} > \max\Big(-\frac{a_1\alpha_4 pr + d_4^*\alpha_1 r + s_4}{\alpha_1\alpha_4 r}, -\frac{a_1 d_4^* pr}{d_4^*\alpha_1 r + s_4}\Big), \quad (56)$$

and therefore we assume that $d_4^*\alpha_1 r + s_4 < 0$. Now, we obtain

$$-\frac{a_1\alpha_4 pr + d_4^*\alpha_1 r + s_4}{\alpha_1\alpha_4 r} > -\frac{a_1 d_4^* pr}{d_4^*\alpha_1 r + s_4}. \quad (57)$$

Thus we see that the condition (56) is equivalent to the conjunction of the conditions

$$\frac{d_4^*}{\alpha_4} > -\frac{a_1\alpha_4 pr + d_4^*\alpha_1 r + s_4}{\alpha_1\alpha_4 r} \text{ and } d_4^* < -\frac{s_4}{\alpha_1 r}. \quad (58)$$

The inequality

$$\frac{d_4^*}{\alpha_4} > -\frac{a_1\alpha_4 pr + d_4^*\alpha_1 r + s_4}{\alpha_1\alpha_4 r}$$

is satisfied if and only if

$$d_4^* > -\frac{a_1\alpha_4 pr + s_4}{2\alpha_1 r}.$$

Finally, we obtain the condition (55). ■

Proposition 6. *If $d_4^* < 0$, then A_0 has negative coordinates. If*

$$0 < d_4^* < \frac{\alpha s_4(s_2\beta_1 - a_1 d)}{d(a_1 d - \alpha_1 s_2)}, \quad (59)$$

then A_0 is a stable node. If

$$d_4^* > \frac{\alpha s_4(s_2\beta_1 - a_1 d)}{d(a_1 d - \alpha_1 s_2)}, \quad (60)$$

then A_0 is a saddle.

Proof. Now we study the stability of the stationary points as solutions to (15). Assume that $d_4^* > 0$. The Jacobian for the point A_0 has only real eigenvalues. The eigenvalues $\lambda_2 = -\alpha s_4/d_4^* - d$, $\lambda_3 = -d$ and $\lambda_4 = -d_4^*$ are always negative. Hence the stability of the point A_0 depends on the sign of the first eigenvalue

$$\lambda_1 = a_1 - \alpha_1\frac{s_2 d_4^*}{d d_4^* + \alpha s_4} - \beta_1\frac{\alpha s_2 s_4}{d(d d_4^* + \alpha s_4)}. \quad (61)$$

If $\lambda_1 < 0$, then A_0 is a stable node. If $\lambda_1 > 0$, then A_0 is a saddle. We check the sign of λ_1 depending on the value of d_4^*. Consider the auxiliary function

$$f(d_4^*) = a_1 - \alpha_1\frac{s_2 d_4}{d d_4 + \alpha s_4^*} - \beta_1\frac{\alpha s_2 s_4^*}{d(d d_4 + \alpha s_4^*)}. \quad (62)$$

The derivative of this function is always positive. Therefore the function $f(d_4^*)$ is strictly increasing and is equal to zero if

$$d_4^* = \frac{\alpha s_4(s_2\beta_1 - a_1 d)}{d(a_1 d - \alpha_1 s_2)}.$$

■

The stability analysis for the point A_2 in the case of Eqns. (15) is the same as for Eqns. (14). Thus we do not repeat it.

Corollary 3. *If one of the conditions (54) or (55) is satisfied, then A_2 is a saddle or a stable point with all nonnegative coordinates.*

While comparing the two models, we should draw our attention to the fact that the case described by (14) seems to be safer for the patient. Even the administration of a high dose of vaccine does not cause a loss of the stability of the point A_0. In the next part of our paper, using numerical simulations, we try to answer the question which type of immunotherapy leads to a quicker cancer remission.

6. Numerical Simulations

We perform numerical simulations for the models (11) and (13). First we intend to present the way in which we obtain some parameters. In our simulations we consider one day as the most natural time unit.

We assume that the time of one mitosis of a cancer cell is between 48 and 72 hours depending on the malignancy of the cancer (Villasana, 2001). On this basis we can estimate the range of the cancer proliferation rate. Thus we obtain $a_1 \in [0.23, 0.35]$.

T helper cells make up about 28% of all lymphocytes presented in peripheral blood[3] (Traczyk, 1997). Since in 1 mm^3 of the blood of a healthy individual there are about 2500 different lymphocytes, we obtain that there are about 700 T helper cells in w 1 mm^3. We assume that the initial number of T helper cells is 700. The lifetime of these lymphocytes is very fluent, sometimes it may be even equal to 10 years, but on average it is 5 years (Traczyk, 1997). Under this assumption we can compute that the death coefficient of T helper cells is $d_4 = 0.00055$. Of course, in the case of the lack of foreign antigens the concentrations of the lymphocytes are on balance. Thus we obtain $s_4 = 0.38$ (because $s_4/d_4 = 700$).

NK lymphocytes make up from 10% to 30% of all lymphocytes presented in the peripheral blood and the medium range is about 15% (Jakóbisiak, 1995; Traczyk, 1997). But the number of NK cells increases with age.

[3] All the quoted values are estimated and concern peripheral blood.

For example, for persons with the Down syndrome ageing is much quicker than for healthy individuals and in these patients the cancer disease is more frequent (Jakóbisiak, 1995). The cancer occurs mainly in old persons and hence we assume that NK lymphocytes make up 20% of all lymphocytes presented in peripheral blood. Therefore, we obtain that in $1\,\mathrm{mm}^3$ of blood in healthy persons there are about $2500 \cdot 20\%$, that is, 500 NK cells. In homeostasis in peripheral blood where there is only a small number of LAK cells, we assume that the initial value of LAK lymphocytes is 1.

The determination of parameters s_2 and d is more difficult, because the proper lifetime of NK cells is unknown (Jakóbisiak, 1995). We assume that it is 2 years (Michałkiewicz, 2003) and hence we obtain $d = 0.0014$ and $s_2 = 0.68$.

The parameters β_2 and α_4 describe the cancer immunogenity. We assume that their values belong to the interval $[0, 0.05]$ (Kirschner and Panetta, 1998). We also assume that the parameters α_1, β_1, α and q_4 are equal to 0.00001, 0.001, 0.0001 and 0.0004, respectively.

Before considering the effects of the treatment methods referred to in the above neoplastic process, let us see how the immune system reacts when it identifies cancer antigens but does not receive additional support in the form of vaccine.

Figures 1 and 2 show the numbers of the particular types of lymphocytes and cancer cells when the patient does not receive treatment. In both cases it was assumed that the number of cancer cells at the time when the immune system identified their antigens is 1000 per $1\,\mathrm{mm}^3$. After having recognized the tumor antigens, the organism is able to overcome the tumor if the tumor does not proliferate too quickly, as presented in Fig. 1. However, in the case when the tumor proliferates very quickly, the organism cannot recover by itself even if the immunological system recognizes the foreign antigens (as presented in Fig. 2).

Let us now study how the population of cancer cells changes if the patient's immune system is supplemented by the investigated vaccine. From now on we make our numerical simulations for the cancer with a proliferation coefficient equal to 0.35.

Figures 3 and 4 show two simulations of cancer evolution when a patient is locally administered a high dose of T helper cells activated outside the patient's body. Our simulations indicate that when the patient receives vaccine in the form of adoptive immunotherapy administered in a continuous manner, it may overcome the tumor.

Performing the simulations of cancer evolution when the patient is in active immunotherapy, we assume that the coefficient s_2 is constant and equal to its physiological value 0.38. Now we change the value of parameter d_4^*. We assume that the administered vaccine may cause an increased proliferation of T helper cells, by shortening the time of the interphase between two consecutive mitosis phases, but the vaccine cannot make the proper time of mitosis shorter.[4] Therefore the absolute value of d_4^* cannot be greater than 0.6.

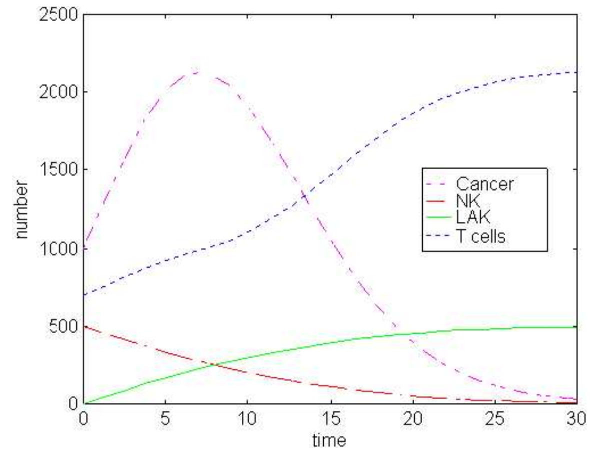

Fig. 1. Simulation of the cancer behavior for $a_1 = 0.23$.

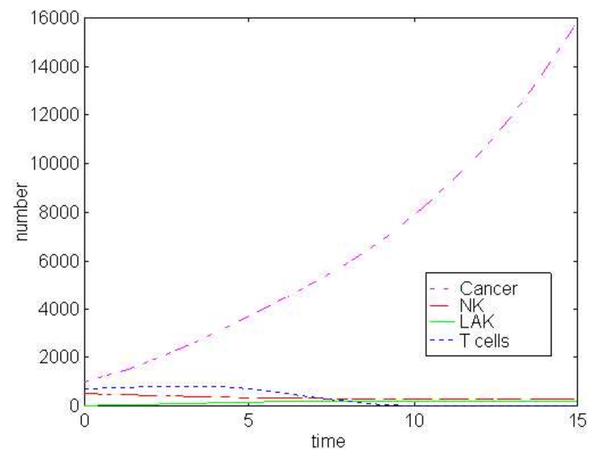

Fig. 2. Simulation of the cancer behavior for $a_1 = 0.35$.

Figures 5 and 6 show two simulations of the cancer behavior when a patient is vaccinated to simulate the proliferation of T helper cells. The vaccine is administered in a continuous manner. The simulations show that a proper application of active immunotherapy leads to a recovery, although the final result strongly depends on the right choice of the vaccine level. In Fig. 5 one can see the simulation of the cancer behavior when the vaccine level is not properly choosen. Although the cancer is finally eliminated, it is not satisfactory that before this hap-

[4] The time of one cycle of mitosis is about thirty hours.

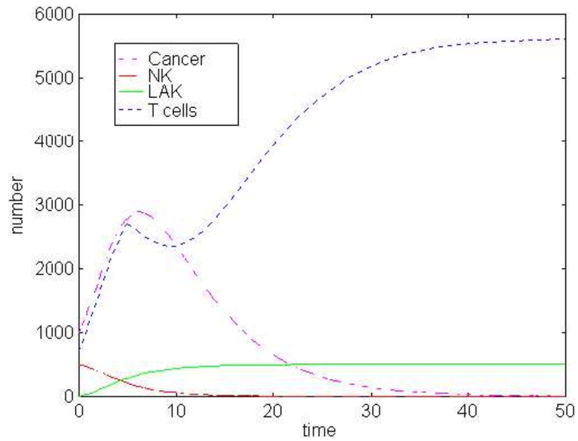

Fig. 3. Simulation of the cancer behavior when adoptive immunotherapy is applied through a continuous five-day administration of a high dose of helper T cells activated outside the patient's body ($s_4^* = 380$).

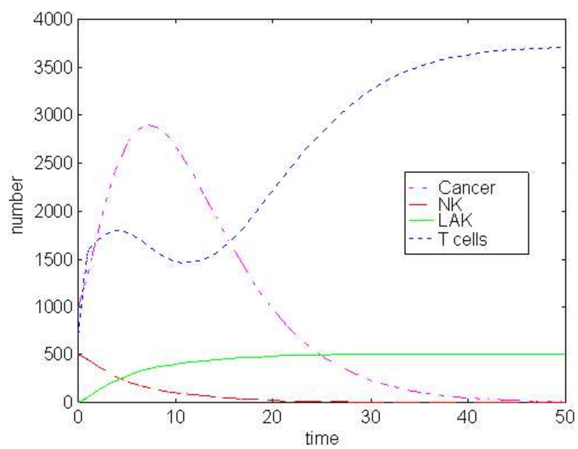

Fig. 4. Simulation of the cancer behavior when adoptive immunotherapy is applied through a continuous one-day administration of a high dose of helper T cells activated outside. The patient's body ($s_4^* = 760$).

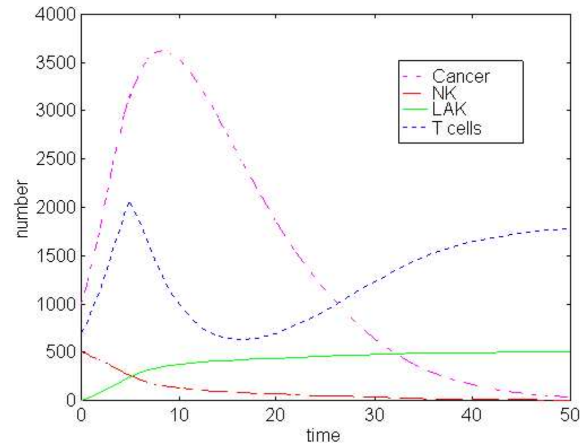

Fig. 5. Simulation of the cancer behavior when active immunotherapy is based on a five-day administration of vaccine stimulating the proliferation of T helper cells ($d_4^* = -0.2$).

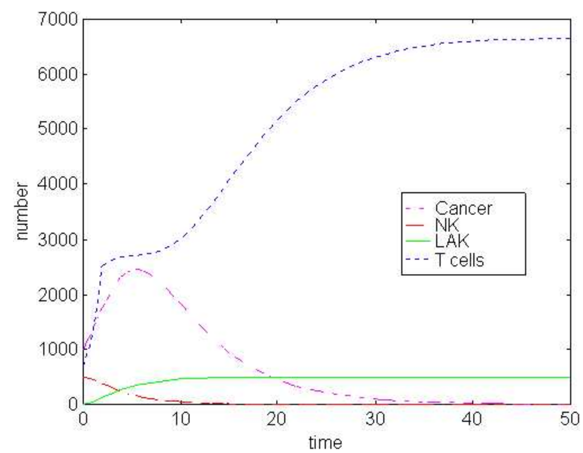

Fig. 6. Simulation of cancer behavior when active immunotherapy is based on a two-day administration of vaccine stimulating the proliferation of T helper cells ($d_4^* = -0.6$.

pens there are far too many cancer cells which may cause metastasis.

A problem related to the treatment methods described by us is the frequently significant increase in the number of T helper cells in the final period of treatment. Although the number of these lymphocytes returns to a normal level in time, this occurs after a very long period (Fig. 7).

The simulations conducted by us suggest at least a partial solution to this problem. If the number of T helper cells administered in the vaccine is increased and the duration of the infusion is shortened, the subsequent increase in the number of these lymphocytes is significantly lower (Figs. 8 and 9).

7. Conclusions

The question arises which of the analysed treatment methods is better, i.e., which of them is more effective in destroying the cancer and which is safer for patients. On the basis of the results of all simulations it was found that the optimal method of cancer treatment is the one leading to a rapid rise in the level of T helper cells, but in which the duration of administering the vaccine is as short as possible so as not to cause an excessive rise in the population of T helper cells that may endanger the patient. Cytokines produced by T helper cells are toxic also for normal cells. A high number of T helper cells causes a high concentration of cytokines and may lead to self intoxication.

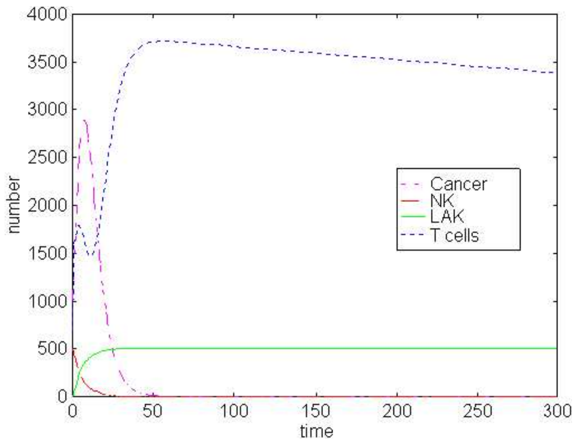

Fig. 7. Simulation of the cancer behavior when adoptive immunotherapy is applied through a continuous five-day administration.

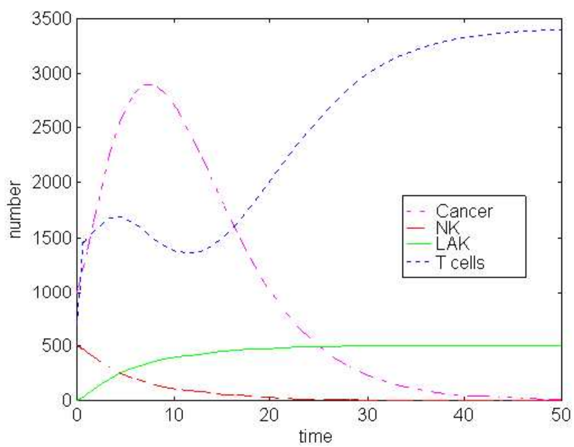

Fig. 8. Simulation of the cancer evolution when adoptive immunotherapy is applied through a continuous fifteen-hour administration of a very high dose of T helper cells activated outside a patient's body ($s_4^* = 1140$).

The analysis of the simulations shows that the method of adoptive immunotherapy seems to better fulfil the hopes inspired by immunotherapy. When comparing the effectiveness in terms of killing cancer cells, this type of immunotherapy is presumably safer for the patient since it causes the smallest increase in the number of other lymphocytes. The simulations also show that the course of active immunotherapy is highly dependent on the activity of the vaccine and the proper time of its administration (Figs. 5 and 6). The advantage of the adoptive over the active immunotherapy can be seen by comparing Figs. 8 and 9.

The main problem in applying the presented models is the determination of parameters used in these models. It is difficult to obtain even such data as the average lifetime

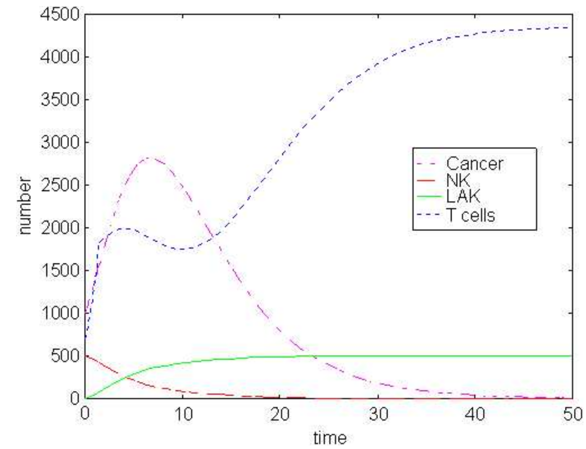

Fig. 9. Simulation of the cancer evolution when active immunotherapy is based on a one-and-a-half-day administration of vaccine stimulating the proliferation of T helper cells ($d_4^* = -0.6$).

of the particular types of lymphocytes or the intensity of immune reactions in healthy subjects, not to mention data for cancer patients. It should be emphasized, though, that this is primarily because most of the necessary parameters are difficult to measure.

The models should be improved to better reflect the cancer evolution and its therapy using vaccine. It seems to be a good modification to replace the square terms describing the stimulations of individual groups of lymphocytes by a function in the Michaelis-Menten form (Eqns. (5)–(7)). This type of kinetics was considered in the paper (Mayer *et al.*, 1995). Such changes would bring our models closer to reality.

Another good modification of the presented models can be extending them by including T suppressor lymphocytes, which are the lymphocytes responsible for the regulation of the immune response. Taking them into account could reduce the disadvantageous increase in some lymphocytes in the last part of treatment. We should mention here that such modifications would probably make the stability analysis of stationary points impossible.

Although the presented models do not show all the complicated physiological and patophysiological processes, by describing their crucial factors they approach the future methods of practically using the vaccinations in cancer therapy.

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