Computer Modeling of Brain Tumor Growth

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Abstract: An important objective of brain tumor modeling is to predict the progression of tumors so as to provide guidance about the best possible medical treatment to halt or slow the tumor's growth. Such computer models also provide a deeper insight into the physiology of tumors. In addition, one can study various what-if scenarios, for instance, investigating the response of tumors following the administration of a drug or a variety of drugs. Abrupt changes in growth rate can also be important for surgical decision-making. Despite increased interest in modeling techniques, relatively little progress has been made in improving such technologies. One problem is the limited data available from patients, typically 1 to 3 MRI (magnetic resonance imaging) sessions, from which one has to extrapolate the type of tumor so as to successfully predict its evolution over time.

Here, the biological and clinical aspects of tumor growth and treatment with surgery, radiotherapy and drugs are discussed in the light of a patient with a brain tumor showing accelerated growth over time. Then, the contributions of mathematical modeling of tumor growth and effects of treatment are presented. Current tumor growth models can be roughly divided in three main categories, (i) cellular and microscopic models that emphasize isolated cell behavior, (ii) macroscopic models that concentrate on the development of cell density over time, and (iii) hybrid approaches that contain elements of both microscopic and macroscopic models. The mathematical theory that underlies these simulation methods is remarkably similar to the physical theory that forms the basis of protein modeling and molecular mechanics tools. A severe limitation of current models is that they are in fact ***not*** patient-specific at all.

INTRODUCTION

A 36-year-old male enters the hospital due to blackout and amnesia. MRI scans reveal a relatively small but malignant brain tumor. The man is eventually diagnosed with anaplastic astrocytoma which is operated radically the following day. The patient recovers from surgery but later develops slight clumsiness in his left leg and arm. He receives standard radiotherapy and CCNU treatment. Some residual tumor is seen on the MRI scan, and because of the tumor's growth and worsening symptoms, the patient is operated again 2 years 4 months after the first operation. After macroscopically total tumor removal, the residual cell growth accelerates and the patient is operated for the third time only 7 months after the second operation. His clinical outcome allows for this operation and he feels better after the last operation. However, the tumor progresses and enormous growth is seen on the MRI shortly after the third operation. The patient's clinical condition also deteriorates and he is hospitalized. Tumor growth continues uncontrollably and the patient dies at the age of 40 years, over four years after the first operation.

Brain tumors are the leading cause of cancer death in children and the second most common cause of cancer death in young adults, and they account for a significant proportion of cancer deaths in older adults [1, 2]. The above patient case exemplifies the typical progression of malignant brain tumors and the difficulties of treating such tumors successfully. Brain tumors consist of a wide range of tumors of which gliomas are the most common. These infiltrative tumors can be divided into low and high grade according to their malignancy [3, 4]. Low grade gliomas are graded I-II and they are usually referred to as diffuse astrocytoma, oligodendroglioma and mixed glioma but the group also includes pilocytic astrocytoma, ependymoma, pleomorphic xanthoastrocytoma, subependymal giant cell astrocytoma and ganglioglioma [5]. Anaplastic astrocytoma, anaplastic oligodendroglioma (grade III) and glioblastoma multiforme (grade IV) are the most typical high grade gliomas.

Most neoplasms arise from a single altered cell with the progeny of that cell expanding as a neoplastic "clone" [6]. In the case of brain tumors this clone usually grows in two ways. At first, the cells simply proliferate and increase tumor core mass, but then at some subsequent time cells become infiltrative and they invade surrounding normal brain tissue [7]. In order to grow, a tumor requires nutrients that are supplied by the vasculature. After a tumor exceeds a few millimeters in size, it requires the production of new blood vessels (angiogenesis) to meet its metabolic demands. Other factors influencing glioma growth are hypoxia and apoptosis.

Tumor progression means that tumors often become more aggressive and more malignant, although the time course may be quite different depending on the histological characteristics of the tumor [8]. Gliomas have the capacity to invade surrounding normal brain which usually prevents complete removal of the tumor. Therefore, malignant recur-

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rence may also represent an aggressive component of the original tumor that was missed on the first resection [9].

The time to recurrence and the type of the recurrent tumor are highly variable and, in general, cannot be predicted from the histopathological and clinical characteristics of the primary tumor [10]. Low grade astrocytomas often recur and progress to higher malignancy. Some astrocytomas show no change in histological grade over more than 10 years following the first operation, whereas others show a rapid transition to malignancy within 1-2 years. However, progression from anaplastic astrocytoma to glioblastoma occurs within approximately 2 years [9, 11].

This review will discuss various aspects of brain tumor growth and will provide an overview of simulation models for tumor growth. *In silico* approaches to study various biological systems and processes have become very mature. Various scientific disciplines have developed their own models and approaches, and in fact general theories of modeling and simulation have emerged [12]. In particular the importance of mathematical modeling to study cancer has been recognized [13]. This review is organized as follows. First an overview of the factors that affect brain tumor growth is provided. This is followed by a review of theoretical methodologies that can be employed for the study of brain tumors, supplemented with a number of representative examples. The description of theoretical models is limited to those methodologies that describe one or more of the factors possibly affecting brain tumor growth. The focus is on concepts, rather then on describing every recent development for each individual methodology. The review closes with a short discussion and an outlook on further developments.

BRAIN TUMOR GROWTH

A malignant brain tumor represents an extremely complicated system and its behavior is affected by a number of factors and (chemical) processes. Among these angiogenesis and hypoxia, necrosis and apoptosis, and motility and invasion of neighboring tissues, are the most important ones. We present below a brief discussion of each of these so as to provide an overview of the life cycle of a tumor and a background for the simulation models.

The patient case introduced above provides a typical, although rarely so well documented, example of the growth of malignant gliomas, originally an anaplastic astrocytomas (WHO grade III, see below). Therefore, it serves as a background for this review. The life cycle of this particular tumor is illustrated in Fig. (**1**). At the outset based upon contrast-enhanced MRI, this temporo-parietal subcortical tumor is about 4 cm in diameter Fig. (**1a**). The resection is radical leaving only a small part of the tumor behind Fig. (**1b**). Due to the location of the tumor the patient develops a weak paresis in his left leg and arm after the resection. He also receives radiotherapy (RT) and chemotherapy (CCNU) after the resection due to the small residual and the nature of the infiltrative astrocytoma. The tumor starts to grow slowly again and 18 months after the resection growth can be seen with MRI Figs. (**1c** and **1d**). The growth continues and tumor resection is scheduled Fig. (**1e**). Prior to the operation the patient suffers again from worsened paresis of his left hand, but his overall clinical condition is good.

The postoperative MRI scans shows that the tumor bulk was removed macroscopically totally Fig. (**1f**). But the diffusive nature of astrocytic tumors leads to growth again. This time growth is accelerated and soon the tumor size overtakes the size before the second operation Figs. (**1g** and **1h**). The tumor has also progressed to glioblastoma multiforme (grade IV). The patient status before the third operation is fairly good; he is able to walk despite the left hemiparesis. He recovers well again after the operation and the left hemiparesis has partly improved. The operation is partial because the tumor has grown to areas which cannot be surgically resected Fig. (**1i**). Shortly after the third operation (2 months) the tumor progresses to a very large size Figs. (**1j** and **1k**) and therefore cannot be surgically treated anymore. The patient's condition deteriorates rapidly as tumor growth has become uncontrollable and the patient dies four and half years after the first surgery.

Angiogenesis and Hypoxia

Angiogenesis, the formation of new blood vessels from an existing vasculature, has been proven to be essential for the growth and expansion of tumors [14,15]. It is a complex and dynamic process which is regulated by several pro- and anti-angiogenic proteins. Among the pro-angiogenic factors (also termed mitogens, as they stimulate cell division) are for instance the vascular endothelial growth factor (VEGF) [16] and the basic fibroblast growth factor (bFGF), while common anti-angiogenic factors are for example angiostatin and endostatin [15, 17]. Angiogenesis requires new capillaries sprouting from existing blood vessels as well as endothelial precursor cells [18].

It has been widely accepted that most tumors originate as small avascular masses [14] that induce the development of new blood vessels once they grow to 1-3 mm^3 in size [19]. However, recent *in vivo* studies have identified an early stage in glioma growth that involves the recruitment of normal cerebral vessels before actual angiogenesis [20, 21]. Experiments suggest that the following steps are involved in tumor growth: perivascular organization, proliferation, apoptosis and vascular involution or regression followed by necrosis and angiogenesis [21, 22].

At some point, as the tumor grows, the normal tissue cannot support further growth and the tumor cells at the center become necrotic as a consequence of a combination of hypoxia (nutritional deficiency) and increased mechanical pressure [23]. This in turn stimulates the tumor mass to release pro-angiogenic factors (also termed cytokines) that diffuse into the surrounding tissue, thus creating a concentration gradient of pro-angiogenic factors. The pro-angiogenic factors ultimately reach the endothelial cells of nearby blood vessels, triggering a series of events that eventually leads to the formation of new vessels that grow towards the tumor and subsequently penetrate it resulting in vascularization of the tumor. The up-regulation of VEGF in glioblastoma multiforme (GBM) is mediated by HIF (hypoxia-inducible factor) transcription factors, which under condition of hypoxia display an increased binding to a number of oncogenes [17]. The involution of host vessels also leads to hypoxia, which in turn induces VEGF release leading to angiogenesis [21].

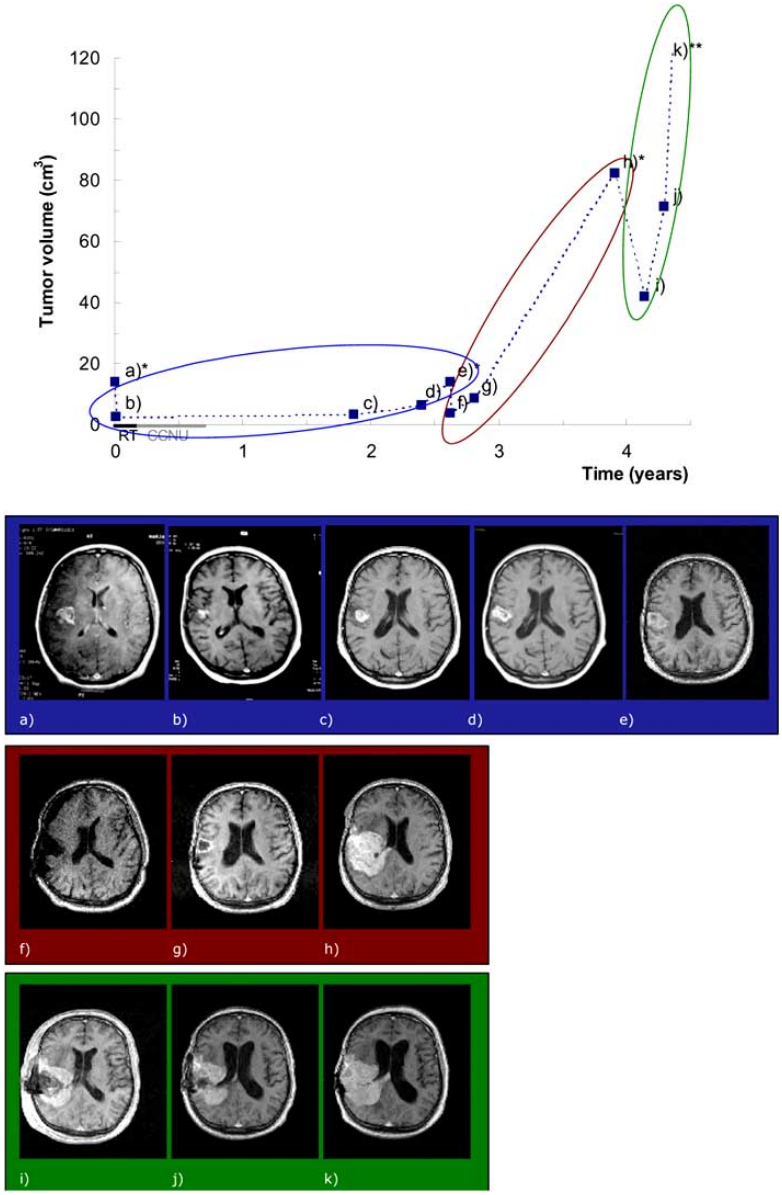

Fig. (1). A typical growth curve of a malignant brain tumor with pertinent MRI scans at 11 points of time. The patient has received both chemo- and radiation therapy (CCNU and RT) after the first surgery and underwent through total of three surgeries (*). Last measurement shows an accelerated growth with tumor volume contrast enhanced mass over 120 cm^3 (**). See the text for additional details. (Marin and Koivukangas, Unpublished data).

The new vessels grow in the direction of the tumor as a consequence of the initial chemotaxis of endothelial cells up the concentration gradient [24-27]. The establishment and maintenance of a sufficient vessel network are required for growth and expansion of normal and neoplastic tissue [28]. The tumor stimulates capillary proliferation which, in turn, promotes tumor growth.

Apoptosis and Necrosis

Tumor tissue growth depends on cell proliferation and apoptosis. These two processes occur simultaneously with tumor suppressor gene, p53, regulating their complex relationship. As the tumor grows, cell proliferation often outweighs apoptosis [29, 30].

Apoptosis is a highly regulated and energy-dependent form of cell death that is usually referred to as programmed cell death. Apoptosis gives rise to a distinct morphology of dying cells [31]. In contrast to apoptosis, necrosis is a passive form of cell death that results from acute cellular injury [32].

Apoptosis significantly influences the growth rate of a tumor and reflects genetic changes [33]. In astrocytic tumors a positive correlation between apoptosis and tumor grade has been reported [34] but the relationship between spontaneous apoptosis and glioma grade is not as straightforward. In glioblastomas, cell death occurs predominantly by the process of necrosis rather than by apoptosis. Consequently, necrosis is one of the glioblastomas' histological features [35].

Motility and Tumor Cell Invasion

Normal glial cells *in vitro* and probably *in vivo* are motile. This property is enhanced in brain tumor cells enabling these cells to invade the CNS (central nervous system) [36]. Cell migration appears to follow white matter tracts; the corpus callosum and anterior commissure are among the major pathways for spread of astrocytomas. These two routes allow the tumors to spread from one hemisphere to the other. Individual glioma cell migration tends to follow anatomical structures like basement membranes of blood vessels and glial limitans externa that contains extracellular matrix proteins, or along myelinated fiber tracts of white matter [37].

Gliomas are very invasive by nature. They are known to grow at a wide range of rates, but little is actually known about these rates or the extent of diffusion or infiltration into the surrounding brain [38]. Because of their natural motility and tendency to infiltrate or diffuse to surroundings, gliomas are rarely cured by surgical resection alone. Adjuvant therapies, including the administering of new drugs, are common ways to treat such tumors.

There are generally three main factors that affect the spread of gliomas cells in the CNS, namely certain anatomical barriers, passive cell displacement, and active cell movement [39]. Specific anatomical and also biochemical barriers prevent tumor cells from metastasizing out of the CNS. It has been shown that leptomeningeal cells and associated acellular components could act as a barrier for glioma cells, thereby preventing brain tumor cells from entering blood vessels, although cells from other types of cancers may pass [40]. Passive cell displacement is thought to be accommodated by the flow of the cerebrospinal fluid within the brain. To what extent this contributes to the actual spread of tumor cells is still unclear.

Active cell movement involves the interaction of cell surface receptors with numerous extracellular matrix molecules (such as fibronection, a major component of the extracellular matrix enhancing the adhesion of cells to the matrix, consequently affecting their ability to diffuse through the matrix), the secretion of proteases, cell signaling events, and the cytoskeleton. Active cell movement can be divided into three general components: protrusion of the leading edge of the cell, adhesion of the leading edge and deadhesion at the cell body and rear, and cytoskeletal contraction to pull the cell forward [41, 42].

WHO Grading System

To facilitate the classification of tumors[1], the World Health Organization (WHO) has introduced a grading scheme [3, 43], where tumors are categorized according to their microscopic appearances. The scheme employs the mitotic index (growth rate), the vascularity (blood supply), the ability to invade neighboring tissues, the resemblance to normal cells, and the presence of a necrotic center as the classification criteria. The resulting grade serves as a measure for the level of malignancy.

In this scheme, grade I tumors represent the least malignant tumor. These tumors grow slowly and microscopically appear almost normal. Although surgery alone may be effective, if it is inaccessible for surgery even a grade I tumor may be life-threatening. Grade I tumors are often associated with long-term survival. Grade II tumors grow slightly faster than grade I tumors and have a slightly abnormal microscopic appearance. These tumors may invade surrounding normal tissue, and may recur as a grade II or higher grade tumor.

Grade III tumors are malignant. These tumors invade surrounding tissue and contain actively reproducing abnormal cells. Grade III tumors frequently recur, often as grade IV tumors. Grade IV tumors are the most malignant and most problematic type of tumor. They have a great ability to invade wide areas of surrounding normal tissue. These tumors reproduce rapidly, appear very unusual microscopically (they exhibit histological signs of very rapid grow) and are necrotic at the center. Such tumors stimulate new blood vessel formation (angiogenesis) to support their rapid growth.

It should be noted that malignant tumors may in fact consist of several grades of cells. Usually the most malignant grade of cell found determines the overall grade of the tumor.

SIMULATION MODELS

The development of theoretical models to study brain tumor *in silico* has been a very active field. Numerous models and simulations methods have emerged over the years. Current models to simulate the growth of brain tumors generally fall into three categories:

- Microscopic models that emphasize the discrete nature of cells.
- Macroscopic models that concentrate on the evolution of cell density over time and space.
- Hybrid or multiple scale approaches that contain elements of both microscopic and macroscopic models.

Single Cell Migration

Cell motility and migration play a critical role in a large number of biological systems, including cancer where metastatic cells may migrate from the tumor mass to other locations [41, 42, 44]. The highly migratory behavior of the infiltrative astrocytomas, a highly problematic brain tumor that ranges from low to high grade, makes them very difficult to resect, because there almost always remains a small fraction of tumor cells in the brain such that the tumor eventually progresses to higher grades [17, 45, 46]. However, the exact details of how tumor cells interact with the parenchyma remain elusive.

A number of modeling approaches that could provide insight into the determinants of migration concentrate on individual cell motility (see, for instance, references [47-50]). Such a description is appropriate in the case of the aforementioned astrocytomas where individual tumor cells percolate through the central nervous system's parenchyma [17]. A recent contribution to this field was made by Zaman

[1] This section has been adopted from http://www.irsa.org/overview.html.

et al. [51, 52]. Most, if not all, other approaches are concerned with movement on two dimensional (2D) surfaces ('crawling'). Instead, Zaman *et al.* have modeled the active movement of a single cell in a three dimensional (3D) matrix, which is more typical for cells. In the context of the topic of this work, the matrix would represent the parenchyma and the mobility considered is governed by haptotaxis. Chemotaxis was not considered in Zaman's model.

Zaman *et al.* assume a stochastic approach, motivated by experimental data, so that a cell will display a random walk-like behavior. The total force $\mathbf{F}_t$ acting on a single cell when the cell is moving through the matrix is composed of three contributions,

$$\mathbf{F}_t = \mathbf{F}_{drag} + \mathbf{F}_{protrusion} + \mathbf{F}_{trac} . \quad (1)$$

Here, the drag and protrusion force (the first two terms on the right, respectively) act as the viscous (frictional) and random force as they also do in the Langevin equation [53]. The drag force models the resistance of the matrix to cell motion, which is in turn caused by other forces acting on the cell (protrusion and traction, the latter being the third term on the right of Eq. (1)). This drag force is assumed to be proportional to the current velocity of the cell and depends on the viscosity of the matrix, a parameter of the model. The direction of the protrusion force, causing an extension of the cell in the direction of the force, is considered to be entirely random and is caused by actin polymerization [41, 42, 51, 52].

The traction force in Eq (1) is composed of two opposing forces, which model the 'force per ligand-receptor complex' at the trailing and leading ends of the cell, which depend in turn on Young's modulus (a measure for the stiffness of material) of the matrix. In addition, the traction forces depend on 'adhesivity', a measure of the strength of the binding between the receptors on the cell membrane and their ligands in the surrounding matrix.

While the model of Zaman *et al*. is simple indeed and possibly too general, it does succeed in providing qualitative insight into the process of cell movement and motility, an extremely complicated process. A key observation was that matrix stiffness strongly affects the cell motility, in addition to surface adhesion and tractile forces that are also apparent in 2D. Also steric factors, proteolysis (to allow migration through steric barriers where the pore size is significantly smaller than the cell dimension) and cell morphology play a role. An adapted version of such a model could therefore be employed to study the specifics of tumor cell motility in brain matter.

Cellular Automaton Models

While the focus of the previous section was on a single cell, a basic starting point for cellular automaton (CA) models is that tumors are considered as a collection of interacting self-organizing cells. Simulation models based upon CA can provide insight into the collective behavior of cells, *i.e.* phenomena that cannot be studied or explained by single cell models, such as those of the previous section.

In CA models of brain tumors, cells are modeled as the lattice sites[2] of a large 2D or 3D lattice. Cells are therefore considered as discrete entities. Each lattice site has or is in a particular state which could be as simple as 'dead', 'alive' or 'vacant' (the latter indicates that the lattice site is currently not occupied by any tumor cell) [54], but more elaborate schemes that mimic the cell life cycle more closely are of course possible and have in fact been implemented (*e.g.* see references [55-57]. The growth of the tumor is modeled by letting cells undergo cell division in which the daughter cell will occupy a neighboring vacant lattice site.

More formally, the excellent review by Moriera and Deutsch [58] defines CA models as a class of spatially and temporally discrete dynamic systems that undergo a variety of local interactions. They provide the following (adapted) list of main characteristics of CA:

1. *Discrete space*: the accessible space is entirely defined by a regular lattice. The number of lattice sites is fixed. The lattice spacing and the number of sites define the length scale of the simulation.
2. *Discrete state*: Each lattice site has or is in a particular state. The state is possibly composed of a finite number of sub-states, each referring to a particular aspect of a real tumor cell.
3. *Discrete dynamics*: The simulation is performed in a number of discrete steps, each corresponding to a fixed time interval (time step). The total number of time steps defines the time scale of the simulation. The update of the system is synchronous (that is, all sites are updated simultaneously).
4. *Local rules*: The evolution of the system is according to given transition rules. These are local rules in the sense that the dynamics of each lattice site depends only on the states of its immediate neighbors (*e.g.* the ability of a cell to produce an offspring through cell division depends on whether there is a neighboring vacant site.)
5. *Homogeneity*: No site is different from any other site. The transition rules apply to each site in the same way at all times.

While these characteristics seem very rigid, it is quite possible to relax or to extend them. For instance, it is not at all required to allow only synchronous updates and it is certainly permissible to implement different transition rules for different lattice sites representing for instance different types of tumor cells.

Düchting [54] appears to be one of the first who has applied lattice models to simulate the response of a collection cells to certain events, such as the sudden removal of a portion of the cells so as to model surgical removal of a tumor. During these early attempts, computing power was not readily available and consequently the lattice sizes were necessarily limited (*e.g.* a 10×10 lattice was employed by Düchting). Currently, lattices with dimensions of for instance 200

[2] The literature also commonly refers to lattice sites as lattice 'cells', 'elements' or 'nodes'. In order to avoid confusion with biological cells, the term lattice site is employed in this review.

x 200 or 100 × 100 × 100 are rather common. These dimensions seem modest, but they are compatible with the number of atoms in for instance molecular dynamics simulation of proteins, where it is feasible to simulate tens of thousands of atoms [59, 60].

An interesting example was recently provided by Kansal *et al*. [61]. These authors rely on 3D lattice model for tumor growth where the lattice is in fact not regular at all but is represented by a Delaunay lattice derived from a Vorronoi network (a collection of polyhedra), as illustrated in Fig. (**2**). The simulation protocol only allows cells that are relatively close to the surface to proliferate; cells located at the interior (center) are considered to be necrotic (this implies that the transition rules are not local). A nutrient gradient is imposed over the tumor as well so as to mimic diffusion limitations in necrotic regions of the tumor. Simulations were initiated from just a few cells allowing the tumor to pass through a multicellular tumor spheroid (MTS) until a macroscopically identifiable size is reached. This size is defined by a parameter that is controlled by pressure responses due to an expanding tumor. The model produces a tumor that displays a growth quite similar to that predicted by the classical Compertz function (see next section).

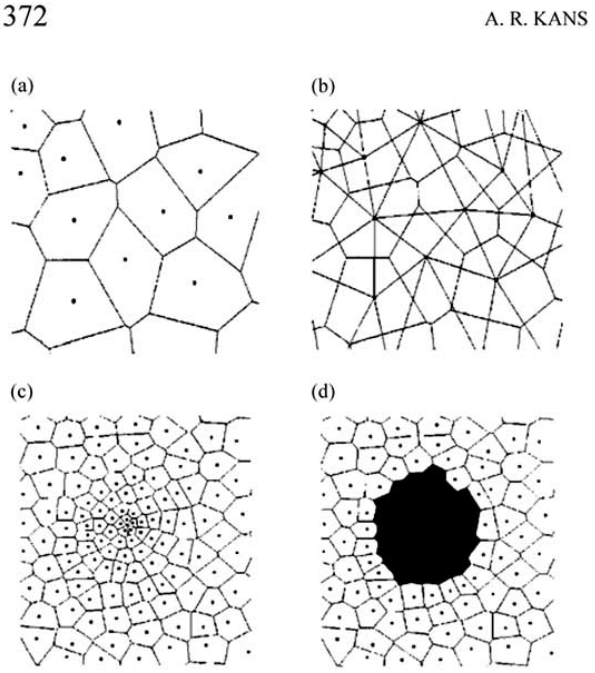

Fig. (2). Illustration of a 2D space lattice tiled into Voronoi cells. The dark portions represent lattice sites occupied by a tumor cell. (From reference [61], reprinted with permission from Elsevier, Copyright Elsevier. All rights reserved).

CA-based models can easily be extended to introduce for instance a second tumor cell type with different growth parameters. As malignant tumors often consist of a number of distinct subclonal populations, Kansal *et al.* [62] further expanded the original model to consider different tumor subpopulations with different behavior and properties. The same CA model was also extended to simulate the important effects of angiogenesis during the early stages of brain tumor growth [63].

Macroscopic Models

Cellular automaton models are limited by the length scales that they can accommodate. This is an obvious consequence of the limited size of the lattice. While it is quite possible to model tumor cell invasion into neighboring tissues, if the tissue is included in the lattice, it is generally not feasible to model tumor cells that spread to other regions of the brain. Generally, the processes by which tumor cells spread inside the central nervous system are extremely complex [39]. Brain tumors rarely metastasize outside the central nervous system, but malignant brain tumors are characterized by diffuse infiltrative growth such that tumor cells can in fact diffuse to very different regions of the brain (as exemplified in Fig. (**1**)). This is in particular true for glioblastoma multiforme (GBM), the most aggressive form of gliomas arising from glia or their precursors [17, 46].

Continuum models of tumors concentrate on the average behavior of cell density instead of emphasizing the discrete nature of cells, while also considering the larger scale of cell behavior [68, 64-67]. The mathematical model is based upon a combination of a diffusion and a growth term (frequently termed the reaction-diffusion equation),

$$\frac{d\rho(\mathbf{r},t)}{dt} = -\nabla \cdot \mathbf{J} + \frac{\partial \rho(\mathbf{r},t)}{\partial t}. \quad (2)$$

This equation assumes that the *total* change $\frac{d\rho(\mathbf{r},t)}{dt}$ of the cell density (concentration) $\rho(\mathbf{r},t)$ at position (location) **r** and time *t* is governed by a diffusion (chemokinesis) term (first term on the right) and a proliferation (growth) term. Here, **J** is the flux of cells which is assumed to follow the phenomenological equation $\mathbf{J} = -D\nabla\rho(\mathbf{r},t)$ or Fick's first law of diffusion [68].

Eq (2) subsequently becomes

$$\frac{d\rho(\mathbf{r},t)}{dt} = D\nabla^2\rho(\mathbf{r},t) + \frac{\partial \rho(\mathbf{r},t)}{\partial \mathbf{t}}. \quad (3)$$

Here, *D* is a diffusion constant which is a parameter of the model. In its simplest form, the brain is assumed to be a homogeneous medium through which cells are diffusing so that *D* has the same value everywhere. This term simply assumes that cell migration is a Brownian-like random motion, an assumption very similar to that of Zaman *et al.* [51]. Typical values for *D* are in the range of 10^{-3} to 10^{-2} mm^2 day^{-1} [65, 67]. The exact value depends in fact on the material, *e.g.* glioma cells migrate about 5 to 100 times faster in white brain matter than in gray [69]. It is quite possible to obtain independent estimates for *D* from simulations that concentrate on single cell migration (see the section on *Single cell migration* above), although recent estimates have resulted in lower values of *D* in comparison to those obtained from the observed motion of a moving glioma front [38].

The proliferation or growth term, the second term on the right of Eq (2), models the evolution of cells due to cell division. This is commonly represented by a simple first order differential equation of the form,

$$\frac{\partial \rho(\mathbf{r},t)}{\partial t} = f(\rho(\mathbf{r},t)). \quad (4)$$

Several choices of f have been employed [67],

$$f = k\rho \quad \text{(Exponential proliferation)}, \quad (5)$$

$$f = k\rho \frac{\rho - \rho_m}{\rho_m} \quad \text{(Verhulst or Logistic Law)}, \quad (6)$$

$$f = k\rho \ln\left(\frac{\rho_m}{\rho}\right) \quad \text{(Compertz Law)}. \quad (7)$$

The factor k in Eqs (5) to (7) is a parameter with units of a rate constant. For the simplest approximation (exponential growth), it specifies the relative increase of cell density or concentration per unit time; a typical value is 0.0012 day^{-1} [38, 65, 67]. A typical outcome of a simulation based upon Eq (2) is shown in Fig. (**3**). Note the strikingly similar appearance of Figs. (**1**) and (**3**) (they are not identical; Fig. (**1**) shows a subcortical peripheral tumor growing centrally, whereas in Fig. (**3**) the tumor grows from a central location peripherally).

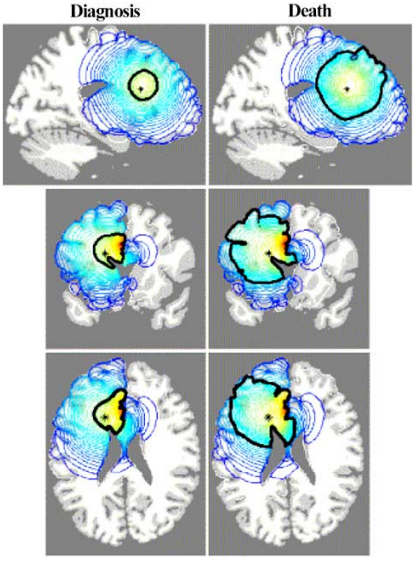

Fig. (3). A typical outcome of a computer simulation of a truly three dimensional virtual brain tumor. The calculation is based upon a continuum approach. The thick black line defines the edge of the detectable tumor by MRI. The left column refers to the moment of diagnosis (the tumor's dimension is about 3 cm), while the right shows the moment of death (the tumor's dimension is about 6 cm). Clearly observable is the density beyond the detectable edge of the tumor, emphasizing the highly diffusive nature of this tumor. (From reference [65], reprinted with permission from Nature Publishing Group, Copyright Nature Publishing Group. All rights reserved).

The Compertz Law for tumor growth has been and still is a very popular model to simulate the growth of a population of cells. Eqs (4) and (7) can be combined to give

$$\frac{\partial \rho}{\partial t} = k\rho \ln \rho_m - k\rho \ln \rho = \alpha\rho - \beta\rho \ln \rho \,. \quad (8)$$

The first term on the right refers to the actual growth (the parameter α is a growth rate constant), while the second is a decay term (the parameter β is a decay rate constant). The Compertz Law is appropriate for tumor cells in the *absence* of migration of cells and interactions between cells. A solution of Eq (8) gives a sigmoidal function where the tumor exhibits an initial exponential growth, but slows down at a later stage due to various mechanisms (growth saturation). These mechanisms are not defined by the growth model. The original Compertz Law is a deterministic model. Albano and Giorno [70] have proposed a stochastic model that generalizes the Compertz Law and accounts for possible fluctuations in cell growth. Chignola *et al.* [71] observed time-dependent fluctuations in the volume of a single MTS (multicellular tumor spheroid) and they were able to reproduce this behavior by introducing a white noise term in the classical Compertz Law. Both approaches are examples of a stochastic description of population growth [72].

With the formulation as expressed in Eq (2), it is possible to distinguish tumors according to the ratio k / D [38] that is compatible with the WHO grading system. A high grade tumor (*e.g.* GBM, grade IV) will display high proliferation and also high diffusion, while a low grade tumor (grade I) will display low proliferation and low diffusion. High grade tumors are consequently highly invasive and display a great tendency to migrate to other regions within the brain, so that there are always glioma cells far beyond the observable boundaries of the tumors. Resection of tumor mass has consequently little effect. Very rapidly growing tumors that can be described with a high k / D ratio (low diffusion) are the solid tumors.

Burgess *et al.* [38] argued that the diffusion aspect of gliomas growth is the more important factor in modeling tumors. Diffusion of cells largely depends on the medium through which cells are diffusing. As noted above, the migratory behavior of tumor cells is predominantly through white brain matter; the diffusion in white matter is about 5 to 100 times higher than in gray matter [69]. The reaction-diffusion model can be adapted to this situation by assuming different values for the diffusion coefficient in white and grey matter. It is possible to take this even further by noting that cells preferentially diffuse along the direction of white matter fiber tracts. To accommodate this anisotropy, it is required to consider the diffusion coefficient as a second order tensor instead of a scalar [65, 67].

The basic equation for the continuum approach for tumor growth Eq (2) can be extended to accommodate for instance surgical resection and chemotherapy [65]. Resection is simply modeled by assigning zero densities at selected regions of the brain (usually the core of the tumor), after which the simulation is continued to monitor the tumor's response. The qualitative behavior of real tumors is fairly well reproduced and shows for instance that the reappearance of the tumor is

the highest at the resection boundary. These continuum simulations also confirm the observation that high grade brain tumors cannot be cured by resection alone. As the consequence of chemotherapy is cell death, Eq (2) is extended by a loss term of the form $-G(t)\rho(\mathbf{r}, t)$ where $G(t)$ is representative for the drug(s) taken by the patient. $G(t)$ is usually taken to be a constant. This approach reproduces reasonably well the qualitative behavior of real tumors.

Sander and Deisboeck [73] have proposed a full continuum model that relies on two major processes for invasive microscopic brain tumors where cells undergo motion in response to (1) chemotaxis, caused by the gradient of nutrient concentration, and (2) homotype factor attraction, which in fact is another form of chemotaxis, but is caused by the secretion of a particular chemical compound that attracts other cells. The latter process is similar to the initial event of angiogenesis where pro-angiogenic factors attract endothelial cells. In this full continuum approach, the motion of cells is described by a standard continuum equation, coupled to equations that describe the nutrient concentration (given from diffusion and consumption by cells) and the homotype factor concentration (given from diffusion, production by mobile cells, and decay). Haptotaxis was ignored in the study.

Venkatasubramanian *et al.* [74] proposed a model to incorporate energy metabolism into a growth model of tumors. While not specific for brain tumors, this model is another example of applying continuum equations. The tumor cell population is described by a similar equation as Eq (2), but the model is extended to predict the local glucose, oxygen and lactate concentrations, while also the production and consumption of ATP is accounted for. This model was applied to multicellular tumor spheroids to study the spatial distribution of proliferating, quiescent and dead cells, the latter two of which are strongly affected by diffusion limitations of essential nutrients in tumors.

The 'mechanical interaction' (mass effect) of tumors with invaded tissue can be addressed by simulating brain deformation. Clatz *et al.* [66] proposed to employ regular linear partial differential equations from classical continuum mechanics with the finite element method (FEM) to describe the mechanical behavior of brain parenchyma. In this model, the skull is included as a fixed entity and the parenchyma is assumed to incompressible.

Hybrid Models

The macroscopic approach as introduced in the previous section cannot be adapted to provide information about the fate of individual cells, as in CA-based methodologies. While CA-based methods can model interactions between cells, they are local. They cannot easily incorporate various types of 'long-range' interactions between the various cells (or cell types). Such interactions can occur in the form of certain chemical signals released for instance by necrotic cells at the center of multicellular spheroids, which diffuse through the tumor system and induce a movement of viable cells towards the center of the tumor [55]. Hybrid models combine elements from both macroscopic and microscopic approaches to overcome limitations in microscopic and macroscopic methods and to explicitly describe phenomena that are ignored or cannot be described by these methods.

Dormann and Deutsch [55] introduce a hybrid model that explicitly takes into account mitosis, apoptosis and necrosis as well as nutrient consumption and diffusible chemical compounds (nutrient and a necrotic signal emitted by necrotic cells). The CA lattice in this model is occupied by tumor and necrotic cells. Each lattice site has four 'velocity channels' and one 'resting channel' that can be occupied by at most one tumor or necrotic cell. The velocity channels give cells the ability to move through the lattice according to the 'orientation' (as defined by the velocity channel they currently occupy). The distributions of the chemical compounds to which cells react are modeled by standard diffusion equations. The rates of mitosis, apoptosis and necrosis (modeled by differential equations commonly employed in cell kinetics) at a given point in the lattice depend on the concentration of nutrient. The necrotic signal interferes with the movement of cells (chemotactic motility: cells move in the direction of the lowest concentration of necrotic signal). The model also allows cells to move towards regions with low cell concentration (pressure induced motility). Such a hybrid offers a way to couple important degrees of freedom whose dynamics occur at different length and time scales. Dormann and Deutsch [55] successfully employed this model to study avascular tumor growth of multicellular spheroids.

The recent work of Athale *et al*. [56] links gene-protein interactions at the cellular level with behavior such as migration and proliferation. The intracellular EGFR-mediated signaling pathway that affects both proliferation and migration (and therefore invasiveness) of gliomas cells was modeled as a simplified gene-protein network (Fig. (**4**)) using ordinary first order differential equations to simulate the time evolution of the various molecular species, including glucose and autocrine transforming growth factor (TGFα), the latter activating the EGFR response through phospholipaseC-γ (PLCγ). Cells were simulated as lattice sites of a 2D lattice as in CA and respond to concentration gradients of both glucose and TGFα. Each cell was further divided into compartments allowing for a heterogeneous distribution of molecular species within cells, as for instance accumulation of PLCγ at the leading edge of the cell may affect its migratory behavior. This multiscale model was employed to study the ability of glioma cells to choose between a migrating and a proliferating phenotype, as experiments have indicated that cells never proliferate *and* migrate at the same time, the so-called dichotomy of glioma cells [75]. This basic model was further extended by Zhang *et al.* [57] to account for the effects of hypoxia and to include a more elaborated cell cycle.

A hybrid model to simulate the hugely important process of tumor-induced angiogenesis was recently introduced by McDougall *et al.* [26] (the application itself was not concerned with brain tumors in particular), based upon the earlier continuum model of Anderson and Chaplain [76]. The total change of cell density (endothelial cells) was governed by regular diffusion (first term of Eq (2)), chemotaxis in response to TAFs, and haptotaxis (caused by gradients of fibronectin). The TAF concentration was assumed to follow a simple loss (uptake) term representing some binding of TAF

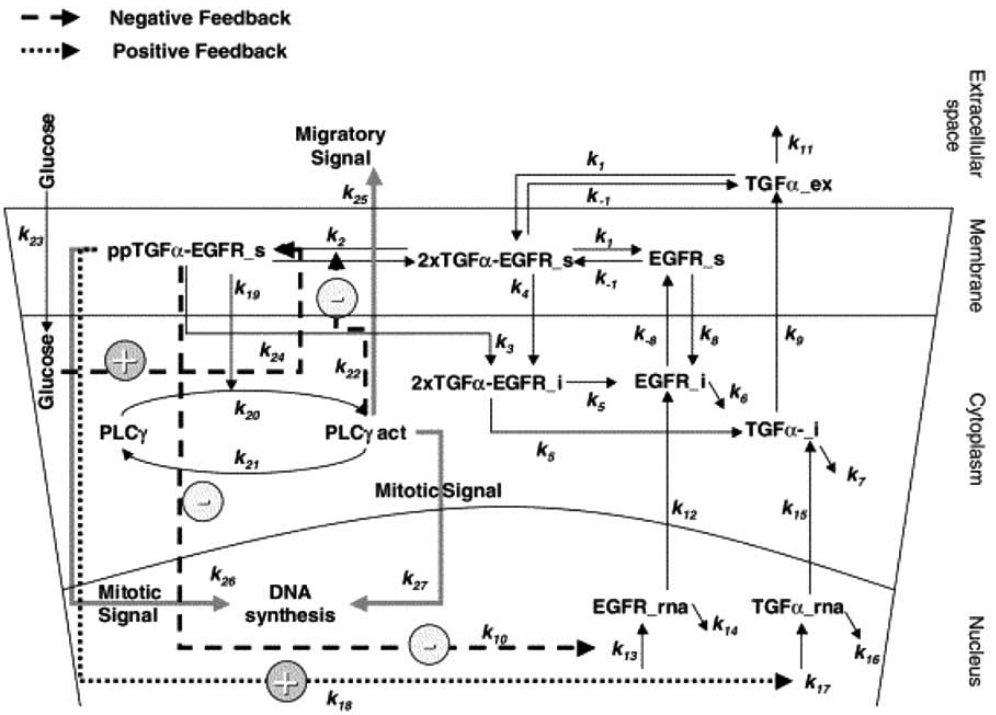

Fig. (4). Illustration of the complex gene-protein network, some of details are given in the text. (From reference [56]. Reproduced with permission from Elsevier, Copyright Elsevier. All rights reserved).

to cells, while the fibronectin concentration was governed by a production (synthesis) and a loss (degradation of fibronection by cells) term. The resulting set of differential equations was solved in a 2D domain. Proliferation of endothelial cells was, however, modeled at a discrete level and in essence corresponded to a biased random walk of individual cells at the sprout tips, allowing for cell proliferation, branching and anastomosis (connections between blood vessels). This model was employed among other things to investigate the flow (blood, drugs) through the vascular network surrounding tumors as a function of for instance blood vessel size, fluid viscosity and blood vessel geometry. Such simulations can be employed for instance to judge the ability of anti-angiogenesis or chemotherapeutic drugs to reach their targets. An example of a simulation that monitors the distribution of a drug in the vascular network is provided in Fig. (**5**).

Alarcón *et al.* [77, 78] introduce a model that is an extension of the hybrid cellular automaton. It accounts for blood flow, structural adaptation of the vasculature, transport of oxygen, interaction of tumor and normal tissue and cell life cycle. These authors strongly emphasize the suggestion that various processes that affect tumor growth take place at different time and length scales and that it is therefore necessary to develop a model that couples these degrees of freedom. They divide their multiple scale model into 'layers', the vascular, the cellular and the intracellular layer, which correspond to the tissue, cellular, and intracellular time and length scales, respectively, as exemplified in Fig. (**6**). As such the models of Alarcón *et al.* [77, 78] and Athale *et al.* [56] share a similar organization in that both models integrate various phenomena occurring at different scales. Alarcón *et al.* employ their model to investigate various aspects of tumor growth such as nutrient heterogeneity and growth laws, and the role of the protein p27 as a prognostic indicator.

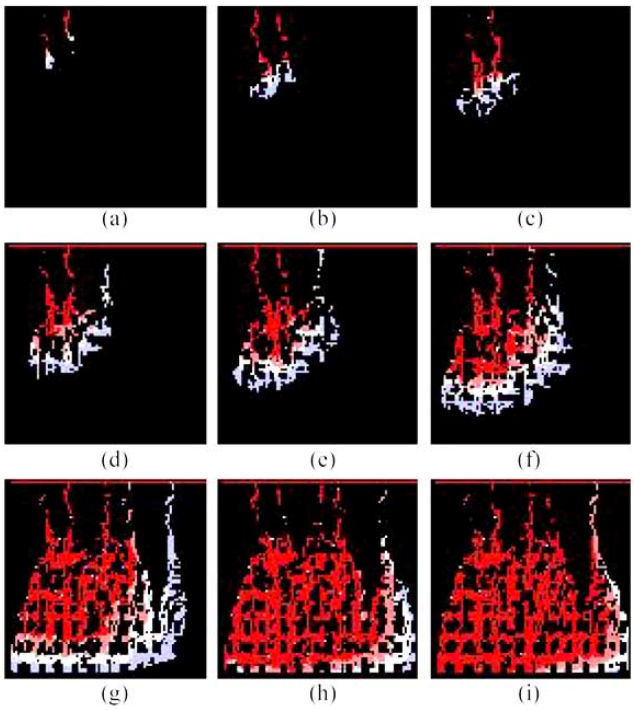

Fig. (5). Drug concentration as it flows from the parent vessel (located at the top of each panel) into the vascular network. The snapshots in panels a to i were obtained at different times in the course of the simulation. (From reference [26], Reproduced with permission from Springer, Copyright Springer. All rights reserved).

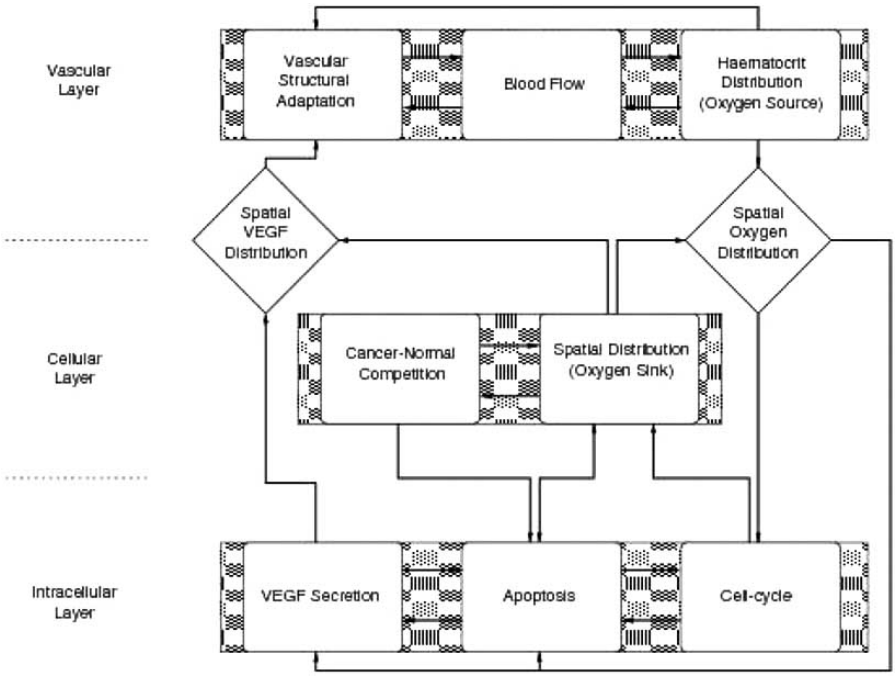

Fig. (6). Illustration of the coupling of events affecting tumor growth at various levels, layers or length scales (From reference [77], Reproduced with permission from the Society for Industrial and Applied Mathematics, Copyright Society for Industrial and Applied Mathematics. All rights reserved).

DISCUSSION

Clinical Impact

Modeling of the growth of malignant astrocytic tumors can be enhanced by examination of individual patients' tumor growth curves. Fig. (**1**) shows the typical development of growth of a representative high grade tumor from a patient series. This patient was operated under intraoperative MRI guidance several times, and after each successive surgical resection, the growth of the tumor accelerated. Also, the effect of surgical resection on tumor size decreased each time.

The effect of surgical resection on a patient's survival is always uncertain. Tumor progression is related to acceleration of the tumor growth. More malignant tumor tissue contains more proliferating cells and new blood vessels provide nutrients for the growth. Individual cell invasion also affects the benefit of such operation because glioma cells diffuse beyond observable boundaries, leading to tumor recurrence after surgical resection. Therefore, therapies that affect motile cells are effective and treatment should be targeted to these cells as well as to the tumor bulk. Theoretical models of tumor growth can be used to predict the growth of a tumor and in theory may have an impact on the decision process to select an appropriate treatment for a given case.

Benefit of Computer Modeling

Computer modeling of tumor growth can be a powerful tool not only to understand why tumors grow as they do, but to provide prognostic estimates of future tumor growth. The effect of new drugs such as temozolomide (Temodal) and of surgical resection can be studied using these kinds of models. Examples of such models were given above.

However, the most interesting aspects of tumor growth modeling lie in three basic questions: First, can the growth models provide retrospective information as to the starting point in time and space when the single tumor cell started to multiply, eventually leading to a clinical tumor? Second, can changes in the rate of growth signal the optimal timing for surgery or other therapy? This would be needed especially in the case of low grade tumors, which are typically stable for years, but then tend to start growing more rapidly at a later point in time, especially if they undergo malignant transformation. Third, is the outcome of a computer simulation specific for a given patient and/or tumor? The first question has not been considered by computer simulation, while the second can be addressed to a certain degree by macroscopic simulations [65, 69]. The identification of the starting point of the tumor in principle could be determined from a brute force approach; many simulations each with different starting conditions, the one resembling the given patient data the best may reveal the starting point. However, one should bear in mind that such macroscopic simulations provide general results at best. The answer to the third question as to whether the simulation outcome truly reflects a given tumor of a given patient has to be negative.

To consider such important questions, and given the fact that tumor growth is obviously an extremely complicated process, very sophisticated simulation models will be required that consider brain tumor growth as a multiple scale process (*e.g.* as in Fig. (**6**)). The latter aspect is of particular relevance since it should be clear now from this review that the events that affect tumor growth take place at the various levels or layers (length scales), are mutually coupled, and occur at different time scales ranging from 10^{-6} seconds to years [56, 77, 78]. For instance, as detailed earlier, the re-

lease of pro-angiogenic factors is triggered by a deficiency of nutrients (such as glucose) which affects the intracellular pathways (intracellular or molecular level, as in Fig. (**4**). These factors diffuse through the tissue space creating a chemical concentration gradient (macroscopic or vascular level) and reaching nearby vessels ultimately stimulating the formation of new blood vessels. These new vessels ultimately provide nutrients to necrotic cells, which may then change their cell state (cellular layer), ultimately stimulating the further growth of the tumor.

Without such a coupling and a sufficiently deep level of detail, computer simulation models for tumor growth are doomed to remain general in nature and will never become patient-specific. The latter concept of predictive individualized care is of particular importance. If such computer simulations models are to become truly relevant in a clinical context, they should be patient-specific to be able to reliably decide whether or not one should operate in a given situation. It also should become possible to predict the chance of success of a certain treatment for a given patient, while at the same time one should be able to distinguish between patients.

Better tumor growth models are also needed to better understand the effects of radiotherapy and new cytostatic and other drugs. Often, the effects of drugs are judged from their ability to bind to given targets (proteins), either through computational approaches [60, 79] or by experiment. While such investigations are important and certainly necessary, the effects of these drugs at the larger scale of tumor development however cannot be inferred from such detailed atomistic investigations and consequently requires a different approach. A number of these 'less-molecular' approaches were briefly explained in this review.

The argument that such models may become too computer intensive and that there still are too many uncertainties in the current understanding of tumor growth are of course valid. For instance, there are many genes involved in cancer, all of which may have a role in the signaling pathway or any other (possibly still unknown) process affecting brain tumor growth. Nevertheless, ongoing and future efforts to develop more sophisticated models for tumor growth should bear the issue of a patient-specific model for tumor growth in mind. The so-called hybrid or multiscale models probably have the highest chance of achieving this goal. The development of more sophisticated models requires further investment in the development of smarter but also more efficient software that take advantage of recent development in scientific computing and improved computing infrastructure (*e.g*., parallel computing, communication protocols between processor cores, dual- or quad- or multi-core central processing units, etc).

CONCLUSIONS

Over the years a number of theoretical models for the simulation of the growth of brain tumors have been developed and applied with various degrees of success. Explicit models are limited by the length and time scale, but can handle many more details, while continuum models can cover large length and time scales but necessarily need to leave out many important details that are relevant to tumor growth. A better understanding of the biological events in tumor growth and progression, coupled with clinical insight, can lead to more comprehensive tumor growth models. It appears that more promising models combine various levels of description into a single model ('hybrid' or 'multiscale' model) for brain tumor growth. Such models should have the best possible chance of becoming truly patient-specific and consequently can play an increasingly important role in the clinical setting.

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ABBREVIATIONS

MRI	=	Magnetic resonance imaging
CCNU	=	A chemotherapy agent
RT	=	Radiotherapy
VEGF	=	Vascular endothelial growth factor
bFGR	=	Basic fibroblast growth factor
CNS	=	Central nervous system
CA	=	Cellular automaton
GBM	=	Glioblastoma multiforme
MTS	=	Multicullelar tumor spheroid
TAF	=	Tumor angiogenic factor

REFERENCES

[1] Mischel, P.S.; Cloughesy, T. Using molecular information to guide brain tumor therapy. *Nat. Clin. Pract. Neurol.,* **2006**, *2*, 232-3.

[2] CBTRUS (Central Brain Tumor Registry of the United States), *2005-2006 Statistical Report: Primary Brain Tumors in the United States Statistical Report, 1998-2002 (Years Data Collected), CBTRUS:* **2006**.

[3] Kleihues P.; Burger, P.C.; Scheithauer, B.W. The new WHO classification of brain tumours. *Brain Pathol.,* **1993**, *3*, 255-68.

[4] Cavenee, W.K.; Furnari, F.B.; Nagane, M.; Huang, H.-J.S.; Newcomb E.W.; Bigner, D.D.; Weller, M.; Berens, M.E.; Plate, K.H.; Israel, M.A. In *Pathology & Genetics. Tumours of the Nervous System*. Kleihues, P.; Cavenee, W.K. Ed.; IARC Press: Lyon, **2000**; pp. 10-21.

[5] Walker, D.G.; Kaye, A.H. Low grade glial neoplasms. *J. Clin. Neurosci.,* **2003**, *10*, 1-13.

[6] Nowell, P.C. Mechanisms of tumor progression. *Cancer Res.,* **1986**, *46*, 2203-7.

[7] Guiot, C.; Degiorgis, P.G.; Delsanto, P.P.; Gabriele, P.; Deisboeck, T.S. Does tumor growth follow a "universal law"? *J. Theor. Biol.,* **2001**, *225*, 147-51.

[8] Foulds, L. Tumor progression. Cancer Res. **1957,** *17*, *355-6.*

[9] McCormack, B.M.; Miller, D.C.; Budzilovich, G.N.; Voorhees, G.J.; Ransohoff, J. Treatment and survival of low-grade astrocytoma in adults--1977-1988. *Neurosurgery,* **1992**, *31*, 636-42.

[10] Hulsebos, T.J.M.; Troost, D.; Leenstra, S. Molecular-genetic characterisation of gliomas that recur as same grade or higher grade tumours. *J. Neurol. Neurosurg. Psychiatry,* **2004**, *75*, 723-6.

[11] Watanabe, K.; Sato, K.; Biernat, W.; Tachibana, O.; von Ammon, K.; Ogata, N.; Yonekawa, Y.; Kleihues, P.; Ohgaki, H. Incidence and timing of p53 mutations during astrocytoma progression in patients with multiple biopsies. *Clin. Cancer Res.,* **1997**, *3*, 523-30.

[12] Zeigler, B.P.; Praehofer, H.; Kim, T.G. *Theory of modeling and simulation*. Academic Press, San Diego (2nd ed), **2000**.

[13] Araujo, R.P.; McElwain, L.S. A history of the study of solid tumour growth: the contribution of mathematical modeling. *Bull. Math. Biol.,* **2004**, *66*, 1039-91.

[14] Folkman, J. Tumor angiogenesis: therapeutic implications. *N. Engl. J. Med.,* **1971**, *285*, 1182-6.

[15] Folkman, J. Fundamental concepts of the angiogenic process. *Current molecular medicine,* **2003**, *3*, 643-51.

[16] Hicklin, D.J.; Ellis, L.M. Role of the vascular endothelial growth factor pathway in tumor growth and angiogenesis. *J. Clin. Oncol.,* **2005**, *23,* 1011-27.

[17] Brat, D.J.; Kaur, B.; van Meir, E.G. Genetic modulation of hypoxia induced gene expression and angiogenesis: Relevance to brain tumors. *Front. Biosci.,* **2003**, *8*, d100-16.

[18] Rafii, S.; Lyden, D.; Benezra, R.; Hattori, K.; Heissig, B. Vascular and haematopoietic stem cells: novel targets for anti-angiogenesis therapy? *Nat. Rev. Cancer,* **2002**, *2,* 826-35.

[19] Vajkoczy, P.; Farhadi, M.; Gaumann, A.; Heidenreich, R.; Erber, R.; Wunder, A.; Tonn, J.C.; Menger, M.D.; Breier, G. Microtumor growth initiates angiogenic sprouting with simultaneous expression of VEGF, VEGF receptor-2, and angiopoietin-2. *J. Clin. Invest.* **2002**, *109*, 777-85.

[20] Holash, J.; Maisonpierre, P. C.; Compton, D.; Boland, P.; Alexander, C. R.; Zagzag, D.; Yancopoulos, G. D.; Wiegand, S. J. Vessel cooption, regression, and growth in tumors mediated by angiopoietins and VEGF. *Science,* **1999**, *284*, 1994-8.

[21] Zagzag, D.; Amirnovin, R.; Greco, M. A.; Yee, H.; Holash, J.; Wiegand, S. J.; Zabski, S.; Yancopoulos, G. D.; Grumet, M. Vascular apoptosis and involution in gliomas precede neovascularization: A novel concept for glioma growth and angiogenesis. *Lab. Invest.* **2000**, *80*, 837-49.

[22] Fischer, I.; Gagner, J.-P.; Law, M.; Newcomb, E.W.; Zagzag, D. Angiogenesis in Gliomas: Biology and Molecular Pathophysiology. *Brain Pathol*., **2005**, *15*, 297-310.

[23] Deisboeck, T.S.; Berens, M.E.; Kansal, A.R.; Torquatto, S.; Stemmer-Rachaminov, A.O.; Chiocca, E.A. Pattern of self-organization in tumour systems: complex growth dynamics in a novel brain tumour spheroid model. *Cell Prolif.,* **2001**, *34*, 115-34.

[24] Vajkoczy, P.; Schilling, L.; Ullrich, A.; Schmiedek, P.; Menger, M. D. Characterization of angiogenesis and microcirculation of high-grade glioma: an intravital multifluorescence microscopic approach in the athymic nude mouse. *J. Cereb. Blood Flow Metab.,* **1998**, *18*, 510-20.

[25] Chaplain, M.A.J. Mathematical modeling of angiogenesis. *J. Neuro Oncol.,* **2000**, *50*, 37-51.

[26] McDougall, S.R.; Anderson, A.R.A.; Chaplain, M.A.J.; Sherrat, J.A. Mathematical modelling of flow through vascular networks: Implications for tumour-induced angiogenesis and chemotherapy strategies. *Bull. Math. Biol*., **2002**, *64*, 673-702.

[27] Chaplain, M.A.J.; McDougall, S.R.; Anderson, A.R.A. Mathematical modeling of tumor-induced angiogenesis. *Annu. Rev. Biomed. Eng.,* **2006,** *8*, 233-57.

[28] Goldbrunner, R.H.; Wagner, S.; Roosen, K.; Tonn, J.-C. Models for assessment of angiogenesis in gliomas. *J. Neuro Oncol.,* **2000**, *50*, 53-62.

[29] Majno, G.; Joris, I. Apoptosis, oncosis, and necrosis. An overview of cell death. *Am. J. Pathol.,* **1995**, *146*, 3-15.

[30] Sarkar, C.; Karak, A.K.; Nath, N.; Sharma, M.C.; Mahapatra A.K.; Chattopadhyay, P.; Sinha, S. Apoptosis and proliferation: correlation with p53 in astrocytic tumours. *J. Neurooncol.,* **2005**, *73*, 93-100.

[31] Kerr, J. F.; Wyllie, A. H.; Currie, A. R. Apoptosis: a basic biological phenomenon with wide-ranging implications in tissue kinetics. *Br. J. Cancer,* **1972**, *26*, 239-57.

[32] Ziegler,U.; Groscurth, P. Morphological features of cell death. *News Physiol. Sci.,* **2004**, *19*, 124-8.

[33] Wharton, S.B.; Hamilton, F.A.; Chan W.K.; Chan, K.K.; Anderson, J.R. Proliferation and cell death in oligodendrogliomas. *Neuropathol. Appl. Neurobiol.,* **1998**, *24*, 21-8.

[34] Bögler, O.; Weller, M. Apoptosis in gliomas, and its role in their current and future treatment. *Front. Biosci.,* **2002**, *7*, e339-53.

[35] Kleihues, P.; Burger, P.C.; Collins, V.P.; Newcomb, E.W.; Ohgaki, H.; Cavenee, W.K. In. *Pathology & Genetics. Tumours of the Nervous System*. Kleihues, P.; Cavenee, W.K., Ed.; IARC Press: Lyon, **2000**, pp. 29-39.

[36] Chicoine, M.R.; Silbergeld, D.L. Assessment of brain tumor cell motility *in vitro* and *in vivo*. *J. Neurosurg.,* **1995**, *82*, 615-22.

[37] Giese, A.; Westphal, M. Glioma invasion in the central nervous system. *Neurosurgery*, **1996**, *39*, 2350.

[38] Burgess, P.K.; Kulesa, P.M.; Murray, J.D.; Alvord, Jr. E.C. The interaction of growth rates and diffusion coefficients in a three-dimensional mathematical model of gliomas. *J. Neuropathol. Exp. Neurol.*, **1997**, *56*, 704-13.

[39] Visted, T.; P.O.; Enger, P.O.; Lund-Johansen, M.; Bjerkvig, R. Mechanisms of tumor cell invasion and angiogenesis in the central nervous system *Front. Biosci.*, **2003**, *8*, e289-304.

[40] Pedersen, P.-H.; Rucklidge, G.J.; Mørk, S.J.; Terzis, A.J.A.; Engelbraaten, O.; Lund-Johansen, M.; Baclund, E.-O.; Laerum, O.D.; Bjerkvig, R. Leptomeningeal tissue: a barrier against brain tumor cell invasion. *J. Natl. Cancer Inst.,* **1994**, *86*, 1593-9.

[41] Becker, W.M.; Kleinsmith, L.J.; Hardin, J. *The world of the cell*, 4th ed., Addison Wesley Longman: New York, **2000**.

[42] Ananthakrishnan, R.; Ehrlicher, A The forces behind cell movement. *Int. J. Biol. Sci.,* **2007**, *3*, 303 -17.

[43] *IRSA,* www.irsa.org, as seen in June 2007.

[44] Lauffenburger, D.A.; Horwitz, A. F. Cell migration: a physically integrated molecular process. *Cell,* **1996**, *84,* 359-69.

[45] Gladstone, C.L. The extracellular matrix of gliomas: Modulation of cell function. *J. Neuropathol. Exp. Neurol.,* **1999**, *58,* 1029-40.

[46] Holland, E.C. Glioblastoma multiforme: The terminator. *Proc. Natl. Acad. Sci. USA,* **2000**, *97*, 6242-4.

[47] DiMilla, P.A.K.; Barbee, K.; Lauffenburger, D.A. Mathematical model for the effects of adhesion and mechanics on cell migration speed. *Biophys. J.,* **1991**, *60*, 15-37.

[48] Dickinson, R.B.; Tranquillo, R.T. A stochastic model for adhesion-mediated cell random motility and haptotaxis. *J. Math. Biol.,* **1993**, *31*, 563-600.

[49] Gracheva, M.E.; Othmer, H.G. A continuum model of motility in ameboid cells. *Bull. Math. Biol.,* **2004**, *66,* 167-93.

[50] Rubinstein, B.; Jacobson, K.; Mogilner, A. Multiscale two-dimensional modeling of a motile simple-shaped cell. *Multiscale Model. Simul.,* **2005**, *3*, 413-39.

[51] Zaman, M.H.; Kamm, R.D.; Matsudaira, P.; Lauffenburger, D.A. Computational model for cell migration in three-dimensional matrices. *Biophys. J.,* **2005**, *89*, 1389-97.

[52] Zaman, M.H.; Trapani, L.M.; Siemeski, A.; Mackellar, D.; Gong, H.; Kamm, R.D.; Wells, A.; Lauffenburger, D.A.; Matsudaira, P. Migration of tumor cells in 3D matrices is governed by matrix stiffness along with cell-matrix adhesion and proteolysis. *Proc. Natl. Acad. Sci. USA,* **2006**, *103*, 10889-94.

[53] Risken, H. *The Fokker-Planck Equation: Methods of Solutions and Applications*: Springer-Verlag Berlin and Heidelberg (2ed new edition) **1996**.

[54] Düchting, W, A model of disturbed self-reproducing cell systems. *Biomath. Cell Kinet.,* **1978**, *2,* 133-42.

[55] Dormann, S.; Deutch, A Modeling of self-organized avascular tumor growth with a hybrid cellular automaton. *In Silico Biol.,* **2002**, *2*, 393-406.

[56] Athale, C.; Mansury, Y.; Deisboeck, T.S. Simulating the impact of a molecular 'decision-process' on cellular phenotype and multicellular patterns in brain tumors. *J. Theor Biol.,* **2005**, *233*, 469-81.

[57] Zhang, L.; Athale, C.A.; Deisboeck, T.S. Development of a three-dimensional multiscale agent-based tumor model: Simulating gene-protein interaction profiles, cell phenotypes and multicellular patterns in brain cancer. *J. Theor. Biol.,* **2007**, *244*, 96-107.

[58] Moreira, J.; Deutcsh, A. Cellular automaton models of tumor development: A critical review. *Adv. Complex Syst.,* **2002**, *5*, 247-67.

[59] Lensink, M.F.; Haapalainen, A.M.; Hiltunen, J.K.; Glumoff, T.; Juffer, A.H. Response of SCP-2L domain of human MFE-2 to ligand removal: Binding site closure and burial of peroxisomal targeting signal. *J. Mol. Biol*., **2002**, *323*, 99-113.

[60] Sharma, S.; Pirila, P.; Kaija, H.; Vihko, P.; Juffer, A.H. Theoretical investigations of prostatic acid phosphatase. *Proteins,* **2005**, *58*, 295-308.

[61] Kansal, A.R.; Torquato, S.; Harsh IV, G.R.; Chiocca, E.A.; Deisboeck, T.S. Simulated brain tumor growth dynamics using a three-dimensional cellular automaton. *J. Theor. Biol.,* **2000**, *203*, 367-82.

[62] Kansal, A.R.; Torquatto, S.; Chiocca, E.A.; Deisboeck, T.S. Emergence of a subpopulation in a computational model of tumor growth. *J. Theor. Biol.,* **2000**, *207*, 431-41.

[63] Gevertz, J.L.; Torquato, S. J. Modeling the effects of vasculature evolution on early brain tumor growth. *Theor. Biol.,* **2006**, *243*, 517-31.

[64] Murray, J.D. *Mathematical Biology*, Springer-Verlag: New York **2002**.

[65] Swanson, K.R.; Bridge, C.; Murray, J.D.; Ellsworth, C.A. Virtual and real brain tumors: using mathematical modeling to quantify glioma growth and invasion. *J. Neurol. Sci.,* **2003**, *216*, 1-10.

[66] Clatz, O.; Bondiau, P-Y.; Delingette, H.; Sermesant, M.; Warfield, S.K.; Malandain, G.; Ayache, N. *Brain tumor growth simulation*, Research report n^0 5187, Institut National de Recherche en Informatique et en Automatique (INRIA): Sophia-Antipolis, France **2004**.

[67] Jbabdi, S.; Mandonnet, E.; Duffau, H.; Capelle, L.; Swanson, K.R.; Pélégrini-Issac, M.; Guillevin, R.; Benali, H. Simulation of anisotropic growth of low-grade gliomas using diffusion tensor imaging. *Magn. Reson. Med.,* **2005**, *54,* 616-24.

[68] Atkins, P.; de Paula, J. *Physical Chemistry*, Oxford University Press: Oxford **2007**.

[69] Swanson, K.R.; Alvored, E.C.; Murray, J.D. A quantitative model for differential motility of gliomas in grey and white matter. *Cell Prolif.,* **2000**, *33*, 317-29.

[70] Albano, G.; Giorno, V. J. A stochastic model in tumor growth. *Theor. Biol.,* **2006**, *242*, 329-36.

[71] Chignola, R.; Schenetti, A.; Andrighetto, G.; Chiesa, E.; Foroni, R.; Sartoris, S.; Tridente, G.; Liberati, D. Forecasting the growth of multicell tumour spheroids: implications for the dynamic growth of solid tumours. *Cell Prolif.,* **2000**, *33*, 219-29.

[72] Barucha-Reid, A.T. Elements *of the theory of Markow processes and their applications*, Dover: Mineola, New York **1997**.

[73] Sander, L.M.; Deisboeck, T.S. Growth patterns of miscroscopic brain tumors. **2002**, *Phys. Rev. E, 66*, 51901- 7.

[74] Venkatasubramanian, R.; Henson, M.A.; Forbes, N.S. Incorporating energy metabolism into a growth model of multicellular tumor spheroids. *J. Theor. Biol.,* **2006**, *242*, 440-53.

[75] Giese, A.; Loo, M.A.; Tran, N.; Haskett, D.; Coons, S.W.; Berens, M.E. Dichotomy of astrocytoma migration and proliferation. *Int. J. Cancer,* **1996**, *67*, 275-82.

[76] Anderson, A.R.A.; Chaplain, M.A.J. Continuous and discrete mathematical models of tumor-induced angiogenesis. *Bull. Math. Biol.,* **1998**, *60*, 857-99.

[77] Alarcón, T.; Byrne, H.M.; Maini, P.K. A multiple scale model for tumor growth. *Multiscale Model. Simul.,* **2005**, *3*, 440-75.

[78] Alarcón, T.; Owen, M.R.; Byre, H.M.; Maini, P.K. Multiscale modelling of tumour growth and therapy: the influence of vessel normalisation on chemotherapy. *Comput. Math. Methods Med.,* **2006**, *7*, 85-119.

[79] Donnini, S.; Juffer, A.H., Calculation of affinities of peptides for proteins. *J. Comput. Chem.,* **2004**, *25*, 393-411.

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