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An evolutionary-game model of tumour–cell interactions: possible relevance to gene therapy

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Abstract

Evolutionary games have been applied as simple mathematical models of populations where interactions between individuals control the dynamics. Recently, it has been proposed to use this type of model to describe the evolution of tumour cell populations with interactions between cells. We extent the analysis to allow for synergistic effects between cells. A mathematical model of a tumour cell population is presented in which population-level synergy is assumed to originate through the interaction of triplets of cells. A threshold of two cooperating cells is assumed to be required to produce a proliferative advantage. The mathematical behaviour of this model is explored. Even this simple synergism (minor clustering effect) is sufficient to generate qualitatively different cell-population dynamics from the models published previously. The most notable feature of the model is the existence of an unstable internal equilibrium separating two stable equilibria. Thus, cells of a malignant phenotype can exist in a stable polymorphism, but may be driven to extinction by relatively modest perturbations of their relative frequency. The proposed model has some features that may be of interest to biological interpretations of gene therapy. Two prototypical strategies for gene therapy are suggested, both of them leading to extinction of the malignant phenotype: one approach would be to reduce the relative proportion of the cooperating malignant cell type below a certain critical value. Another approach would be to increase the critical threshold value without reducing the relative frequency of cells of the malignant phenotype.

Keywords: Cell interactions; Stable clonic heterogeneity; Threshold proportion of cells; Gene therapy; Angiogenesis

1. Introduction

Throughout the development of a tumour, selection of specific mutations is traditionally believed to reflect the ability of these mutations to promote growth or inhibit death by cell-autonomous modes of action [1–4]. An improved understanding of the genetic and biochemical mechanisms, by which mutations confer a growth advantage to cancer cells, has led to the formulation of mathematical models for tumour initiation, growth and metastatic spread [5–9]. These models typically incorporate the effect of putative paracrine and/or autocrine growth factors, but rely nevertheless on the cell-autonomous consequences of mutations in the malignant cells. Recently, Tomlinson and Bodmer [10,11] presented evolutionary-game mathematical models for the time evolution of a tumour cell population which incorporated non-autonomous effects of tumour cell mutations in terms of interactions among cells that have adopted individual genetic strategies. Following the work by Axelrod and Hamilton [12], evolutionary games have been studied as simple mathematical models of the evolution of cooperation in a population. The prototypical example is the Prisoner's Dilemma, where the payoff from a specific strategy depends on whether the other prisoner collaborates or defects. A popular introduction to the Prisoner's Dilemma and other evolutionary games may be found in Nowak and colleagues [13].

Tomlinson and Bodmer discussed several models, and one of these considered the production of a growth factor, exemplified by an angiogenic promoter, that was associated with a cost of production and a replication

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advantage. The benefit was obtained by the cell itself, as well as other cells in the population. As long as the benefit was greater than the cost of production, a stable polymorphism between producers and non-producers were predicted.

2. Materials and methods

Here, an extension of the evolutionary model by Tomlinson and Bodmer [10,11] is proposed and its behaviour investigated. Specifically, this model incorporates a local threshold condition for deriving a benefit from collaboration between tumour cells. One concrete example could be the production of an angiogenic promoter where it is conceivable that producer cells have to collaborate in order to provide a sufficiently high local concentration of the stimulating factor. We describe the simplest type of such a model by introducing a threshold condition for obtaining the fitness advantage, i.e. the unit will only benefit if at least two out of three interacting cells adopt the strategy. In the angiogenic example, this would mean that two of three cells will have to be producer cells before the cells obtain the replication advantage. This mathematically simple model gives rise to rather complex dynamics.

3. Results

3.1. Evolutionary games and tumour population dynamics

Tomlinson and Bodmer [10] developed a mathematical model of the dynamics of a population consisting of two types of cells with strategies A+ and A−. An A+ cell produces a certain factor which conveys a proliferative advantage both to the cell itself and its neighbours. In contrast, the A− cell does not produce the factor and a population of A− cells will only have the baseline reproductive rate. The detailed mechanism of this interaction needs not to be specified, but the resulting net cost or benefit of being A+/A− controls the relative change in the abundance of the A+ phenotype from one generation of cells to the next. Mathematically, each phenotype, in this case A+ and A−, is assigned a 'payoff' or 'fitness' value which weighs the benefits against the costs.

The fitness values are expressed as the baseline reproduction plus the benefit, j, and minus the cost, i (Table 1). The stable equilibrium of A+/A− cell types described by Tomlinson and Bodmer [10] exists as long as $j > i$. Intuitively this is clear. If A+ cells are abundant, it pays to be an A− cell because of the great probability of meeting an A+ cell and thus gaining the proliferative advantage without paying the cost of being A+. This results in the highest fitness ($1+j$). If, on the other hand, the population consists predominantly of A− cells, it pays to be an A+ cell. The fitness pay off ($1-i+j$) of an A+ interacting with A− cells is larger (for $j > i$) than what is gained when A− cells interact [1]. Note that the pay offs are those obtained by the cell type in the top row. Thus, the population will consist of a mixture of A+ and A− cells and their relative frequencies will converge toward a stable internal equilibrium as shown in the mathematical analysis by Tomlinson and Bodmer [10].

The Tomlinson and Bodmer [10] model represents a situation where A+ and A− cells will coexist in a stable genetic polymorphism regardless of the initial frequencies of the two. Thus, in this model, an observed clonic heterogeneity could be explained exclusively by the dynamics generated by the cellular interaction and not by the effect of some epigenetic or external factor.

3.2. The threshold model

The Tomlinson and Bodmer [10] model considers interactions between *two* cellular strategies, with the probability of an interaction expressed as the product of the relative frequencies of A+ and A− cells in the population. Now, assume that a growth promoter produced by the A+ cells needs to reach a sufficiently high concentration before its effect is discernible. This scenario assumes synergistic effects in the interaction among cells. A simple model of this effect assumes that a given cell interacts locally with two cells, and that at least two A+ cells out of the three cells considered are needed to give the cell a proliferative advantage. The 'neighbours' of the cell are drawn at random from the population and the probability of a given interaction is therefore expressed as the product of the frequencies of the cells involved. This simple individual-based metaphor for cellular interaction represents a plausible biological situation where a population-level synergistic effect emerges. The functional relationship between the frequency of A+ cells and the fitness resembles closely that of the Tomlinson and Bodmer [10] model when A+ cell are common. The co-operative effect and synergism matter only in situations where A+ cells are rare. The matrix of fitness parameters is shown in Table 2. Note in particular, that the high pay off is not achieved when one A+ cell meets two A− cells, the A+ cell will only experience the cost, i.

Table 1
Fitness matrix for the Tomlinson and Bodmer model [10]

	Cell	
Neighbour	A+	A−
A+	$1-i+j$	$1+j$
A−	$1-i+j$	1

The frequency, v_{n+1}, of A+ cells in generation $n+1$ is a function of the frequency v_n in generation n, that is $v_{n+1} = f(v_n)$. The transfer function, F, can be shown to be

$$v_{n+1} = f(v_n) = \frac{v\cdot\left[(1-(1-v)^2)\cdot(1-i+j)+(1-v)^2(1-i)\right]}{v\cdot\left[(1-(1-v)^2(1-i+j)+(1-v)^2(1-i)\right]+(1-v)\cdot[v^2\cdot(1+j)+(1-v^2)]}$$

On the right-hand side, the index n has been suppressed to simplify the notation.

3.3. Mathematical properties of the model

The mathematical behaviour of the transfer function above depends on the relative values of i and j. When $j<2i$ all internal equilibria disappear and the frequency of A+ cells goes to zero with increasing generation number. Thus, the A+ property becomes extinct in the population.

A special case is $j=2i$. This creates a single internal point of equilibrium ($v = 1/2$) which is attractive for $v > 1/2$ and repelling for $v < 1/2$.

If $j>2i$, a bifurcation occurs relative to the frequency $v = 1/2$: a stable equilibrium exists above $v = 1/2$ and an unstable equilibrium below $v = 1/2$. The two internal equilibria will change their positions symmetrically as a function of the relationship between the cost and benefit parameters. For j/i tending to infinity the two equilibria tends toward frequencies of A+ cell going to 1 or 0.

Fig. 1 shows a simulation with $j=3i$. The frequency of A+ cells converges towards the stable internal equilibrium or towards zero depending on the initial relative frequency of these cells. The unstable equilibrium acts as a barrier that has to be crossed, in order to change the frequency of A+ cells away from either of the two stable equilibria. The population will reach one of the two equilibria depending on whether the initial frequency of A+ cells is located above or below the unstable equilibrium. In other words, starting above the unstable equilibrium the population frequency distribution will end up in a stable clonic heterogeneity (in population genetics termed a *stable polymorphism*).

Table 2
Fitness matrix for the threshold model

	Cell	
Neighbours	A+	A−
A+, A+	$1-i+j$	$1+j$
A+, A−	$1-i+j$	1
A−, A−	1−i	1

4. Discussion

The most interesting feature of the current model is the existence of an unstable internal equilibrium which forms a barrier between two stable states of the population, one with a stable polymorphism between A+ and A− cells and one where only A− cells are present. During tumorigenesis, it must be assumed that local collaboration is possible which may allow this critical threshold to be crossed locally. At a later stage, the selection pressure against the tumour cells may have increased and this would leave the tumour in the state described by the model analysed here. The model is inspired by the possible effects of spatial heterogeneity, although true spatial structured interactions are not explored here. True spatial effects in a version of the Prisoner's Dilemma game have been thoroughly investigated by Nowak and May [14,15], who showed that spatial models with local effects, compared with panmictic models, can generate qualitatively different results in terms of coexistence or extinction of strategies.

In a therapeutic context, the model suggests the intriguing possibility that gene therapy directed against restoration of tumour-suppressor genes only needs to change the fraction of mutated tumour cells below a certain level, and that once the frequency gets below the unstable equilibrium, internal dynamics will force the mutated cells to extinction. In cancer gene therapy, a great deal of effort has gone into restoring normal p53 function in mutated tumour cells [16]. If immunohistochemical detection of p53 is accepted as an indication of mutations, hotspots of mutated cells have been described [17]. Within these regions, a varying frequency of

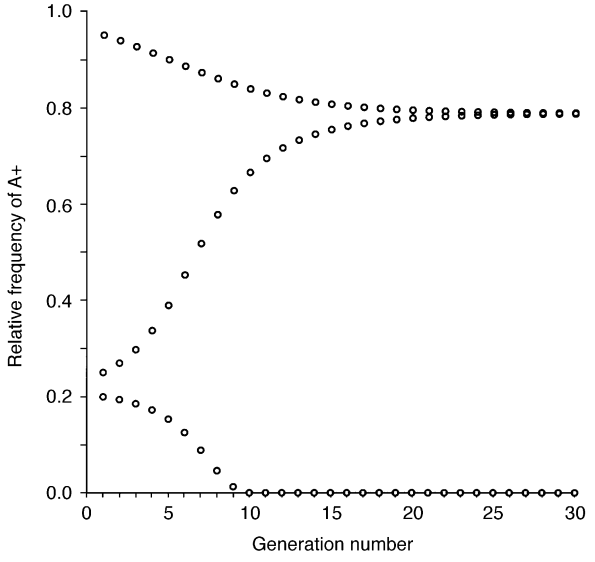

Fig. 1. Simulation with parameters $j=3i$. The ordinate is the frequency of A+ cells and the abscissa is the number of cell generations. Note the extinction of A+ cells when their initial frequency is below that of the internal unstable equilibrium.

positive cells is observed, which is consistent with the evolution of local co-operation between mutated cells. Although current delivery techniques fail to restore p53 function in every single tumour cell, inhibition of tumour growth was observed in the first clinical trial of p53 gene therapy in lung cancer. In mouse models, the therapeutic effect of p53 replacement also exceeds that expected from the fraction of cells expressing the normal *TP53* gene. The *in vivo* observations from clinical trials and mouse models are generally explained by some bystander effect. *In vitro,* an unknown bystander effect requiring intercellular contact has been observed in co-culturing experiments [18]. Alternatively, and entirely in line with the mathematical model presented here, inhibition of the angiogenic response has been proposed as the mechanism for the observed *in vivo* bystander effect seen after restoration of wild-type p53 in only a fraction of tumour cells [19].

In principle, two different therapeutic strategies are suggested from the dynamics of this model. One strategy is to lower the frequency of proliferative active cells (A+) until the ratio of A+ to A− cells gets below the unstable equilibrium. In the example of p53 mutated cells, this ratio can be changed by restoring the normal gene function through gene therapy [20] or by drug-based approaches [21]. The A+/A− ratio can also be changed by selective killing of mutant p53 cells. One such strategy did show promising results in preclinical studies [22], although the specificity for p53-mutant cells in this particular approach has recently been questioned [23]. If the cost and benefit parameters as described here are relevant features of the tumour cell population and can be manipulated, another strategy is to move the unstable equilibrium towards a higher frequency of A+ cells. Thus, by lowering the benefit or increasing the cost, only a very high ratio of A+ to A− cells can maintain a stable equilibrium, and the slightest increase in the frequency of A− cells will lead to extinction of A+ cells. For p53, one possible way to increase the cost could be to induce an immune response against cells expressing mutant p53 [24].

Finally, the model described here on tumour cell collaboration, particularly collaboration on production of angiogenic promoters, could also suggest one factor contributing to the mechanism of sudden metastasis or regression of a tumour. The concept of dormant metastasis, and also the sudden aggressiveness of metastasis following surgical removal of a primary tumour, has recently received renewed attention [25]. One mechanism by which metastasis are believed to be kept in a dormant state by the primary tumour is through secretion of sufficient amounts of angiogenic inhibitors to overcome the angiogenic promoters produced by the metastasis [26]. In the context of the model, a reduction in the amount of external angiogenic inhibitors would lower the threshold condition for obtaining the benefit of producing an angiogenic promoter. In the presence of a primary tumour, i.e. high levels of angiogenic inhibitors, a high threshold level would predict a rare stochastic appearance of metastasis with each proliferating metastasis representing a cluster of cells where the frequency of A+ cells has reached above the internal unstable equilibrium thus allowing a stable polymorphism. This is consistent with a steady rate of mortality from relapsed breast cancer observed for decades after primary treatment. Similarly, lowering the threshold value by removal of external inhibitors is consistent with the sudden increase in the number of proliferative active metastasis occasionally observed following radical primary treatment [26].

If angiogenesis is playing a role in the sudden appearance of proliferative active metastasis, it could also be involved in spontaneous regression of tumours. Indeed, angiogenesis has become a promising target for cancer therapy [27,28]. Interestingly, significant results can be obtained with relatively small changes in the level of angiogenic promoters [29–31]. Czubayko and colleagues [30] observed a marked delay in tumour growth following a 20% reduction in fibroblast growth factor (FGF)-binding protein and concluded that angiogenesis appears to be "exquisitely sensitive" to even small reductions in the stimulatory factors. If threshold conditions for angiogenic promoters exist, the dynamics suggested by the model could be involved in this phenomenon.

5. Conclusion

In the light of recent advances in the description of cell–cell interactions and their biochemical signalling pathways, this kind of model is, in our view, a potentially useful tool for understanding the behaviour of populations of tumour cells. Eventually, these models could lead to more elaborate applications of spatial simulations, where local synergism and clustering effects determine the final composition of the cell population. We propose that some gene therapy approaches may benefit from changes in population dynamics depending on intercellular interactions. This may relieve some of the constraints on current gene-therapy approaches where complete restoration or killing of cancer cells would seem almost unachievable in most human tumours.

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