Six Wedges To Curing Disease

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Abstract. One of the great challenges in ecology and evolutionary biology is to explain disease, whether caused by infectious agents such as parasites and pathogens, or by the deterioration or transformation of cellular behavior and function, a prime example of the latter being cancer. Decades of observation and research suggest that successfully treating disease requires insights into how the environment mediates the interactions between disease causing agents (DCAs) and diseased individuals. A major finding is that single factor, targeted therapies are not only likely to fail in controlling or eradicating many DCAs, but are also likely to select for resistance, reducing options for subsequent treatment attempts, and in cases of infectious DCAs, rendering therapeutic agents (e.g., antibiotics) obsolete.

I argue that meeting the growing challenge of treating disease in agriculture and animal husbandry, in protected and domesticated species, wildlife, and in the human population will require a fundamental understanding of ecological interactions at sites of infection or disease. I discuss different ways in which components of such *disease ecosystems* mediate DCA and therapeutic dynamics and resistance evolution, and derive a very simple mathematical criterion for therapeutic success. I then touch on how fundamental insights as revealed by the processes of evolutionary rescue and competitive release can help understand why therapies succeed or fail. Finally, I present six "wedges" that can each contribute alone, or as part of multi-pronged approaches to successfully treating disease.

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The Magic Bullet

Despite remarkable progress in prevention and treatment, the impacts of diseases in agriculture and animal husbandry, and on protected and domesticated species, wildlife, and human health are likely to intensify into the 21st century. Humans in particular are increasingly in contact with each other and with wildlife, treated with drug regimens that select for resistance, and adopt life-styles or are exposed to environments that render them more susceptible to infectious diseases and more likely to get cellular diseases such as cancers.

For many clinicians the Holy Grail is to discover the Ehrlichian "Magic Bullet"—a drug that will target the disease-causing agents (DCAs) and cure disease. Intuitively, the most effective way to reduce DCAs is to "hit hard and fast". That is, more drug means more kill within the tolerance limits of the patient. Rapid administration of the drug means forestalling further DCA growth and associated symptoms, but also reducing the probability of the appearance of resistant mutant strains. Despite decades of research on pest and disease control (much of it with an ecological basis) this approach and the many alternatives described below remain highly controversial (e.g., [1]).

The Magic Bullet may be shortsighted for another reason. What will cure most patients may not be optimal for the general population in which resistant variants may emerge and spread[1]. This problem is in many ways similar to the objectives of classical pest control [2], where short-term 'success' is to attain economic targets for the treated area (e.g., an agricultural field), subject to meeting the global objective of slowing the spatial spread of resistance [3]. Although human cancers are not transmitted between individuals, the concept from pest control of spatial resistance management could apply to treating metastasis [4], and certain authors have argued that certain targeted therapies could be counterproductive to treatment success [5].

Curing Disease is an Ecological Problem

Some of the diseases that present the greatest threats to human health are caused by microparasites, including viruses, bacteria, and protozoa, and macroparasites such as helminth worms. Microparasites in particular are characterized by very large populations and therefore considerable potential for rapid evolutionary responses. When a drug is applied to a large, diverse population, the response will be determined in part by the absolute (growth) and relative (selection) impacts on sensitive and resistant subpopulations. Given that resistance depends on (epi)gene expression, this means that environment, and more generally ecology, needs to be incorporated into our understanding of why chemotherapies (against microparasites, macroparasites or cancers) succeed or fail.

Disease control within the organism is an ecological problem, but is rarely seen as such. Rather, research and application are overwhelmingly focused on the direct interaction between the therapy (typically a drug) and the DCA. Treatment success means that the drug contributed to clearing the DCA, whereas failure would suggest that the drug choice, dose, and/or schedule was in err, and/or that resistant strains were present. The major omission from this perspective is environment. Indeed, the very same interactions found in terrestrial and aquatic systems are

[1] In the context of infectious disease, transmission and spread are to other hosts, whereas, in cancer (with the exception of transmissible cancers in dogs, Tasmanian devils, and certain bivalve species) spread is either through local or distant metastasis.

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found *within* diseased hosts: intra- and interspecific competition, resource limitation, mutualism, facilitation, and predation.

Here I argue for a framework integrating abiotic and biotic interactions involving DCAs and drugs within *disease ecosystems*. I briefly discuss the basis for this concept. I then present several mechanisms associated with therapeutic failure, all of which involve DCA clonal escape. A criterion for therapeutic success integrating clonal escape is then formalized in a very simple mathematical expression, and I discuss parallels with the concepts of evolutionary rescue and competitive release. Finally, I present six complementary strategies or 'wedges' towards improved control, their ecological basis, and why they should be considered in future theoretical developments.

Table 1. Some basic challenges to treating patients and to disease management over the broader population for several important DCAs affecting the human population.

DCA	Individual patients	Broader population
Bacterial pathogens	Large, diverse populations; hypermutator strains; protective biofilms; multidrug resistance	Horizontal gene transfer of virulence factors and resistance from other bacterial species
Plasmodium	Evades host immune system; resistant strains transmitted to host; distinguishing *Plasmodium* species to determine specific treatment	Difficulty in controlling the vector
HIV	High within-host population turnover and generation of antigen diversity; uses its enemy (white blood cells) to replicate	Prevention through safe-sex and no needle sharing
Cancer	Large, diverse populations; genomic instability; dormancy; refractory cancer stem cells; limited drug amounts due to toxicity; evasion of immune system	

My approach is largely review, and I make reference where appropriate to individual treatment strategies coming from bacterial infections, *Plasmodium*, HIV and cancers. Table 1 presents a list of some of the basic factors associated with these DCAs that make them particularly challenging to treat in individuals and to control at a population level. Comparing and contrasting insights from these different literatures could lead to a greater fundamental understanding of disease ecosystems, how drugs affect them, and translation to clinics.

The Disease Ecosystem

Despite many differences between parasites and tumor cells, one broad similarity is that both live in *disease ecosystems*. The disease ecosystem encompasses birth, growth, survival and inter-individual and inter-populational interactions such as predation (immune system), cooperation (cell-cell signals, tissue architecture, cell function), competition (both direct (as in the case of

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many parasites) and indirect for limiting resources (host cells, glucose, oxygen)), resource replenishment (angiogenesis), detritivory (phagocytosis), and external intervention in the form of therapy. The latter is tempered by the idiosyncratic nature of information gathering and translation into a rational treatment. Related perspectives have recently been discussed for parasites [6], pathogenic bacteria (e.g., [7]), and cancer [8,9].

Figure 1 shows an oversimplified depiction of one type of disease ecosystem: the cancer microenvironment. Tumor growth depends importantly on resource flows through the 3-D mass of uncontrollably dividing, motile cells [10]. Cells that are only a few millimeters distance from the nearest capillary will experience hypoxic stress and lactic acid accumulation, due to deficits in oxygen and glucose. Cells tend to become increasingly hypoxic and anoxic towards the center

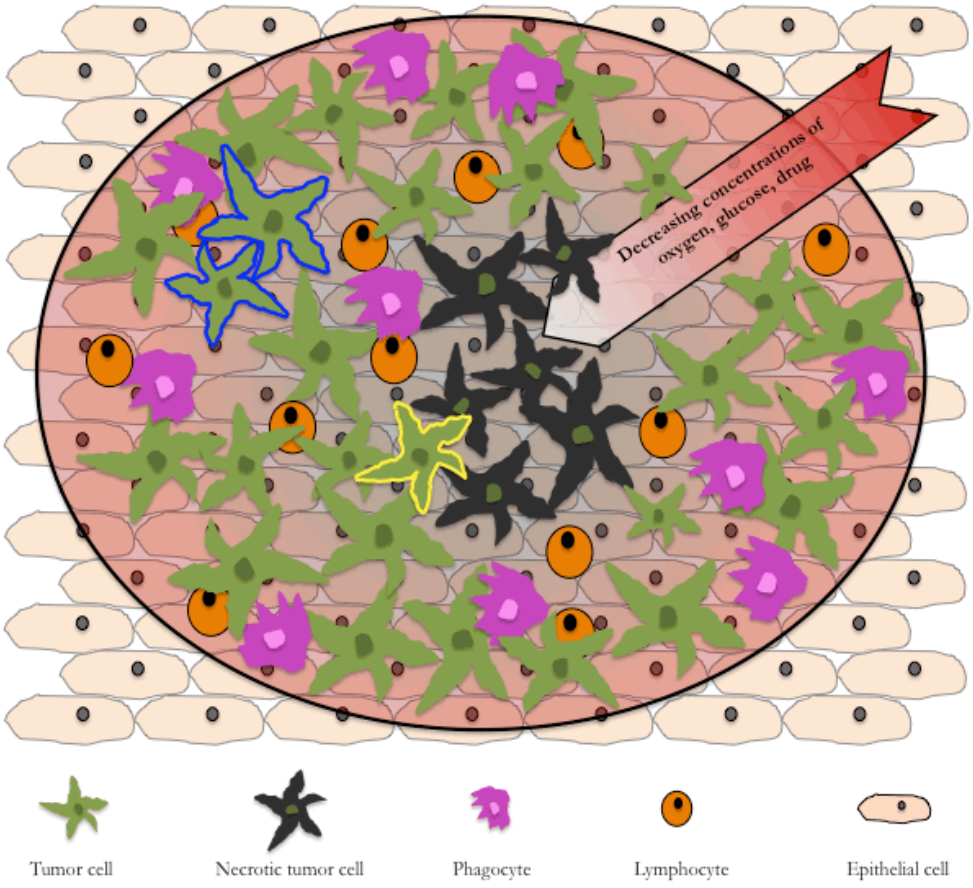

Figure 1. Disease ecosystem for tumor growth. In this oversimplified representation of the tumor microenvironment, tumor cells can grow (green cells) as long as they have sufficient access to oxygen and glucose, and sufficient space. Tumor cells compete amongst each other and with surrounding healthy epithelial cells. The intensity of competition increases with cell-cell proximity and distance from the nearest blood capillary (not shown). In the absence of sufficient resources (toward the tumor core), cells become necrotic (black cells). Tumor cells may be predated by components of the immune response (phagocytes and lymphocytes). Fibroblasts (not shown) modify the disease ecosystem through the production of structural tissue (extracellular matrix) and growth factors, facilitating angiogenesis (pericyte and endothelial cell production), inflammation, immune evasion, tumor growth and metastasis. Tumor cells may escape the immune system (not shown) or therapies, the latter through, for example, genetic resistance (blue outlines) or limited drug contact (yellow outline).

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of a tumor and under prolonged stress they form a necrotic core [11,12]. Stressed cells may send signals to the local vascular system to extend capillaries into the tumor (angiogenesis), which, if successful, promotes further tumor growth [13]. From a therapeutic point of view, the challenge is to arrest angiogenesis whilst promoting conditions for drug diffusion into the tumor (which *increases* with angiogenesis). Those cells experiencing insufficient doses of the drug or in a dormant state (Figure 1, yellow cell) may be the source of future relapse, as are cells receiving the full drug dose, but harboring chemoresistance mutations (Fig. 1, blue cells) (and see below). Little is known about how resource dynamics and cell-to-cell signaling involving DCAs and healthy cells determine the emergence and fates of refuge cells and resistant cells. Finally, the predators in the disease ecosystem are different components of the innate and adaptive immune responses that are most functional at the incipient stages of tumorigenesis, becoming less effective or failing in conjunction with tumor escape [9].

Largely analogous reasoning holds for non-self DCAs (e.g., HIV, bacterial pathogens, *Plasmodium*), though each has its own spatial, behavioral and functional specificities. Moreover, the caricature in Figure 1 is centered on local interactions between DCAs, host cells and tissues, and drugs. A more realistic representation would include regional and global host interactions (e.g., immune, hormonal, other disease foci, impacts on host health and behavior), and in cases of infectious disease, interactions with the external environment. Understanding the differences and commonalities between different DCA ecosystems will form a part of developing theories and predictions for how diseases can be cured.

DCA Escape

Beyond misdiagnosis or misappropriations in the choice of (or patient intolerance to) drugs, the main generic cause for treatment failure is DCA escape. DCAs cause disease, morbidity and mortality in part because they either escape natural host controls or amplify them (e.g., cytokine storms). DCA escape from drugs bears limited resemblance with parasite escape from immune systems or pests escaping natural enemies in biological control. This is in part because drugs are non-adaptive, whereas the body has a wide array of checkpoints for detecting and destroying diseased cells, and the immune system for detecting non-self antigens and the subsequent disabling of parasites.

DCA escape from chemotherapies can occur in one or more of several ways. First, DCAs may produce quiescent or dormant states. Examples include quiescent cancer stem cells [9] and persister bacterial cells [14]. Second, DCAs may be tolerant, whereby mechanisms such as reduced receptor sensitivity or density limit drug impact, as cases of drug tolerant cells in certain cancers [15]. Third, a DCA may directly resist a drug through a molecular change or a change in receptors and/or drug transport. For instance, bacteria may indirectly resist antibiotics through the upregulation of efflux pumps [16], or directly resist via drug modification or inactivation [17]. Fourth, some individual agents may be less attainable by the drug than others, either due to spatial refuges or active escape through plastic behaviors. An example is limited drug diffusion through tumors leading to spatial refuges and resistance [4,18]. Fifth, agents may naturally have high mutation rates, or be induced or selected to have increased mutation rates, such as in hypermutator bacteria [19], HIV [20], and certain cancers [21]. Tumors in particular are thought to contain numerous resistant variants by the time neoplasms are typically discovered [22]. The above five mechanisms are largely inferred from *in vitro* experiments; their detailed roles in treatment failures are largely unknown (e.g., [23]).

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A Simple Criterion

The primary objective of chemotherapy is to impact DCAs so as to durably allay or eliminate disease impacts on health. Outcomes are challenging to predict because the system is potentially very complex, involving how the DCA interacts amongst itself, with other DCAs (e.g., in co-infections) and with healthy cells, but also with other ecosystem components such as resource availability (space, nutrients), habitat quality, the microbiome, and the immune system. The addition of one or more drugs potentially influence these interacting compartments either directly or indirectly, creating possible feedbacks, and in so doing will make predictions based on individual components in isolation, overly simplistic.

The complexity summarized above indicates that simple criteria, such as reducing the DCA net population growth rate below zero, are probably not useful for understanding chemotherapeutic success and failure. Rather, a model incorporating the key phenomenon of DCA escape may provide significant insight into the basic control problem. The model can be modified to incorporate different features of the disease ecosystem.

We assume n distinct asexual, haploid classes of DCA where, for phenotype i, the current population size, maximum growth rate, density dependent limitation (e.g., through competition and/or predation), and reductions in birth (or increases in mortality) from a chemotherapy are respectively $N_{i,t}$, λ_i, $f_{i,t}$ and ϕ_i. Note that ϕ_i encapsulates both the sensitivity of a strain to the drug (given inherent levels of tolerance or resistance), and the eventuality that the strain is also protected due to spatial refuges.

Assuming a large initial population $N_{i,0}$ of the most frequent clone, such that population changes are dominated by deterministic rates, the population at a short time interval Δ into the future is

$$N_{i,t+\Delta} = \lambda_i \phi_i f_{i,t}\{N_{i,t}\} N_{i,t}.$$

Further, assuming that density dependence (intra-population competition) in the DCA becomes negligible soon after the chemotherapy begins, and resistant or protected variants are initially rare, then after a treatment period $x\Delta$, clone i is approximated by

$$N_{i,t+x\Delta} = (\lambda_i \phi_i)^x N_{i,0}.$$

Clearly if $\phi_i < 1/\lambda_i$ for all i, then some level of reduction in density is ensured. The criterion for achieving population control below a target density, T, predicted to result in success is therefore

$$W = \Sigma_i (\lambda_i \phi_i)^x N_{i,0} < T. \quad (1)$$

Importantly, the objective W<T may be initially met, simply because refuge populations are small and do not appreciably grow during the initial phases of the therapy. Should these populations not be sufficiently held in check thereafter, then eventually they will (re)emerge, and potentially falsify the success criterion (1). This may happen should subpopulations escape drug control and/or if the immune system is not sufficiently effective.

Notice the following. First, because this is an exponential population process, the required ϕ to achieve control is highly sensitive to the multiplier λ. This reflects the compounding effect of insufficiently checked exponential growth. Second, meeting criterion (1) requires that *all* genotypes are sufficiently controlled; this sufficiency will depend on the densities, biological

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characteristics of each clone present, and disease ecosystem interactions. Third, if a single resistant mutant is present at the beginning of the therapy, then, should it emerge, it takes a minimum of $\ln(T)/\ln(\lambda_m)$ time units for criterion (1) to be rejected. And fourth, the immune system is an important factor that potentially intervenes to reduce λ, whereby therapy accompanied by immune responses are likely to be more effective than either alone in reducing non-self parasites. In contrast, by the time a cancer is discovered (e.g., poses a health threat), the tumor has likely escaped natural immune control and would require specific immunotherapies to contribute to DCA suppression beyond the effects of a chemotherapy (or radiation therapy).

This model is clearly oversimplified and the inclusion of further realistic positive or negative density-dependent interactions such as competition between tumor cells and surrounding healthy cells could influence the therapeutic outcome. For example, Sottoriva et al. [24] considered the emergence and growth of competing clones in colorectal cancers. They showed that the initial appearance of a set of driver mutations resulted in the dominance of these clones in tumor growth, even when more fit strains subsequently emerged. This is because early emerging clones gain a priority effect (a 'head-start'), and as the population carrying capacity is approached, clonal per capita growth rate and per capita beneficial mutation rate both decrease, resulting in slower clonal turnover.

Rescue and Release

Before proposing a framework towards improved treatment outcomes that incorporates the dynamics of disease ecosystems, I describe two complementary phenomena that are useful in understanding treatment failures.

In *evolutionary rescue*, the effects of a drug drive the sensitive population towards extinction, but resistant variants that are already present or emerge during the treatment 'rescue' the population [25]. The evolution of DCA resistance under high drug doses is a textbook example of evolutionary rescue, though not all cases of resistance evolution need involve evolutionary rescue. Specifically, drugs may only partially impact the sensitive subpopulation due to e.g., DCA plasticity, spatial refuges, or under-dosing. It is also possible that drug resistance evolves in situations where extinction would not have occurred in the first place (e.g., [26,27]). In the context of improving chemotherapies, if the initial DCA population is small and/or genetic variation low, then it may be best to treat with high doses until the DCA is cleared. This assumes that the therapy sufficiently decreases the rate of evolution (i.e., lowers the number of births and it is not mutagenic) and does not induce refuge behavior such as dormancy observed in some cancers [15].

In *competitive release*, populations compete for space or limiting resources, and the reduction of one or more competitors contributes to the growth of other, less impacted (e.g. resistant) populations [2]. Day and colleagues [28] differentiated competitive release from competitive suppression, the latter occurring when one competitor suppresses another through a shared density dependent response, such as an immune reaction or environmental degradation [29]. With a few exceptions, competitive release is not well understood in the context of disease ecosystems. For example, Wargo and coworkers [30] showed an effect of treatment duration on the magnitude of competitive release in *Plasmodium*. Pena-Miller and colleagues [31] used modeling and experiments of antibiotic combination therapies to show that failure by the first antibiotic strongly favors the growth rate and emergence of resistant clones, which, in turn evade control by a second drug. One of the dangers in competitive release (and indeed evolutionary rescue) is that

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the expanding resistant population may evolve compensatory traits [32,33] or acquire other fitness-enhancing traits, such that proliferation potential is comparable or even greater than that of the original population, as has been hypothesized in certain cancer relapses [34].

Ecological and Evolutionary Wedges to Vanquish Disease

As described above, the conventional wisdom in treating disease is to hit hard and fast. The main constraints on dose and duration are toxicity and treatment costs. The risk of this approach is treatment failure and, in the case of infectious DCAs, the spread of resistant strains to other individuals.

Fundamental research aimed at understanding treatment outcomes needs to incorporate the dose debate into a more integrated framework based on the disease ecosystem. I describe six non-mutually exclusive variables and strategies ('wedges') that could contribute to improved treatment outcomes (Table 2).

***1*. Dosing.** Growth inhibition and kill assays are mainstays of pharmacodynamics. However drugs are not static entities once introduced into the body: they are heterogeneously distributed, and may be modified or inactivated, and excreted (pharmacokinetics). For example, Foo and colleagues [35] studied combined evolutionary and pharmacokinetic models of the emergence of cell resistance to erlotinib in patients with epidermal growth factor receptor (EGFR)-mutant lung cancers. They found that pulsed dosing could control tumors with resistant clones, but that sufficiently long treatment holidays risked enabling the emergence of *de novo* resistant clones. Ankomah and Levin [36] employed models of pharmacodynamics, focusing on the dose and duration of antimicrobial therapies in systems with innate and adaptive immune responses. They found that high doses used until the immune response finished clearing the infection were superior to less intense strategies (but see [37] for contrasting findings). Akhmetzhanov and Hochberg [38] used computational models of tumor growth, based on empirical parameter estimates to derive minimal levels of continuous chemotherapy that would extend patient's life expectancy compared to high dose therapies. They found that optimal dosing should no more than counteract the growth of the DCA clone with the highest fitness, otherwise fully resistant clones would likely emerge, resulting in possible treatment failure.

Although there is considerable empirical work on how drug dosing potentially affects treatment outcome (for literature survey, see [37]), it does not provide a clear signal as to whether "hit hard" approaches are superior to alternative dosing schedules [39]. For example, Negri and coworkers [39,40] demonstrated how strains of *E. coli* with different levels of resistance to the antibiotic cefotaxime persisted at different drug doses. They identified the "selective window": doses that eliminate sensitive strains and favor relatively resistant ones. Work on *Plasmodium* has shown sensitivity to treatment dose [29], and Wargo and colleagues [30] demonstrated an effect of treatment duration on the emergence of resistance. A corollary to dosing (intensity, schedule and duration) is whether the therapy should actually kill DCAs (cytotoxic) or rather render them unable to proliferate (cytostatic). Theory shows that these alternatives can have contrasting effects on dynamics (e.g., [41]), suggesting that interactions with other components in the disease ecosystem, such as resource competition or predation by the immune system, may result in different therapeutic outcomes depending on the extent to which therapy kills cells or arrests cells.

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2. **Combination therapies**. Employing two or more therapeutic agents to treat a disease has shown promise for certain cancers [42], bacterial diseases [43] and HIV [44] (see also examples in [45]). The basis is two-fold. First, combination therapies can be devised to foil one or more escape responses, including spatial refuges, effect pathways, and resistance genes (e.g., [46]). The underlying idea is that total population coverage by two or more therapies is greater than any one alone. For example, Komarova and Wodarz [47] parameterized mathematical models to show how the number of drugs and probabilities of resistance to each inform on how many drugs are necessary to successfully treat cancers. Examples of combined approaches also abound in the antimicrobial literature, for example, the use of multiple phage types or combinations of phages and antibiotics against certain pathogenic bacteria ([48,49], but see [50]). Second, the order, schedule, and dose of each anti-DCA can be adjusted for maximal effect [51]. 'Maximal effect' is a complex quantity that integrates the impact on DCA numbers, the probability of resistance, and allaying disease severity, whilst avoiding toxicity issues (especially for cancer therapies).

Scheduling can be an important parameter in achieving maximal effect, for instance when therapeutic agents interfere with one another if administered together. For example, Torres-Barcelo and coworkers [51] showed how the delay between additions of a bacteriophage and an antibiotic is key in maximizing impact and minimizing resistance in populations of *P. aeruginosa*. This is due in part to the density dependent nature of phage multiplication and impact on bacterial hosts. Roemhild and colleagues [52] demonstrated how the order of sub-lethal doses of antibiotics could have substantial effects on bacterial populations and resistance (see also [31]). Combination therapies may also exploit characteristics of the disease ecosystem and of DCAs themselves. Examples include the sequential use of chemotherapy and immunotherapy [53], and polyADP ribose polymerase (PARP) inhibitors capitalizing on synthetic lethality in certain breast and ovarian cancers [54].

3. **Increasing the costs of resistance.** One underexplored approach to improve treatment outcomes is to increase the costs of resistance. The idea is to use specific interventions so that sensitive DCAs competitively control or even eliminate resistant populations. Enriquez-Navas and coworkers [55] have recently argued that for some cancers, chemotherapies could be alternated with "fake drugs" that serve only to tip the competitive balance in favor of sensitive cells. This idea has considerable appeal, but the argument could be made that rapid treatment with a fake drug should be initiated *before* the real chemotherapy is even applied. The reasoning is as follows. The resistant cell population is expected to be at its lowest numbers before treatment, either should resistance have an intrinsic cost relative to sensitive cells, or should it have no cost, but emerged only once the tumor attained large cell numbers. The sensitive cell population will be at its maximum numbers and competitive impact on resistant cells before treatment. Increasing the cost of resistance through a fake drug will further tip the balance in favor of sensitive DCAs. This paves the way for the administration of a bolus of the real drug to clear the infection or the cancer, or if not completely successful, to follow the real drug with cycling fake drug treatments, as in ref. [55].

4. **Dynamic agents.** Drug specificity is one of the mainstays of Ehrlichian magic bullets. However, it is also a shortcoming since, as discussed above, targeting one or a small number of vulnerable traits can select for resistance [5]. An alternative is to capitalize on the diversity of "living" agents and their potential to overcome the evolution of resistance *in situ*, or in treating previously evolved multi-drug resistant pathogens. Examples include lytic phages against pathogenic bacteria [56,57] and oncolytic viruses [58]. For example, Wodarz and Komarova [59]

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developed and analyzed mathematical models to show that tumor control by oncolytic viruses depended importantly on virus growth and diffusion to refuge cancer cell populations. Others have argued for a "Trojan Horse" strategy, whereby avirulent DCA variants could be used to control more virulent strains [60] and greatly reduce the risks of resistance evolution. An interesting related development is to employ bacteriophages as adjuvants to select for increased sensitivity to antibiotics [61].

A prime advantage of dynamic agents, and bacteriophages in particular, is self-amplification and adaptation to resistant hosts *in situ.* Because the diversity of phages is virtually limitless, they can be combined into cocktails to maximize overall strain coverage and impact on the target bacterium [48]. Bacteriophage can either treat established infections, or be used prophylactically ('lying in wait') so as to prevent pathogens from colonizing incisions or wounds [62]. When bacterial densities are low (such that phage self-amplification is limited) or difficult to attain, the phage inoculum can be increased, functioning more like a conventional antimicrobial drug.

***5*. Tweaking different interactions in the disease ecosystem**. As introduced above, one of the cornerstones of ecology—that individuals interact with each other and the environment—is all but neglected in chemotherapies. There are many possible interventions into the disease ecosystem that could increase impacts on DCAs. For instance, immune systems ('predators') can be harnessed to combat malignant cells (e.g., [63]). Empirical work shows that one way to accomplish this is through dynamic agents [64], whereas other immune stimulations prevent DCAs from becoming established in the first place [65]. Other study shows how healthy cells may help suppress neighboring tumors [66], or the evolutionary rates and adaptability of DCAs may be slowed [67], or how factors as diverse as resource bases, virulence factors, host tolerance, microbiomes, and cell-cell cooperation may be targeted to improve therapeutic outcomes [68–70].

***6*. Adapt to the situation.** The conventional approach of treating disease is to employ information based on symptoms, eventual identification of the DCA, and patient characteristics to devise a one-off strategy with the objective of improvement or cure. A different approach is to use early-warning signs, and initial and on-going information to predict disease dynamics and adapt treatments so as to manage or eradicate the DCA. For example, 'adaptive therapy' has the dual objectives of an acceptable cancer burden and minimal resistance evolution (recently reviewed in [71]). In the context of phage therapy, Pirnay and colleagues [56] distinguished general or *prêt à porter* strategies and more personalized *sur mesure* approaches. One example of the latter is adaptive "phage training", whereby lytic phages are evolved before or during a therapy in the laboratory, so as to improve their impact on particular bacterial strains, extend the spectrum of hosts attacked, or anticipate bacterial resistance [72,73] (see also [71] for a related discussion of "steering evolution"). Once training is complete, the trained isolates are then (re)introduced into the disease ecosystem.

More generally, assessing possible 'Plan B' strategies *before* a treatment actually begins is not only sensible, but may also influence the choice of Plan A. Forecasts of ecology and evolution in the disease ecosystem should guide such choices. For example, the use of a risky Plan A with no Plan B should Plan A fail (e.g., the treatment of a multidrug-resistant bacterium with a last resort antibiotic), may be inferior to the employment of a less aggressive, slower-acting Plan A (the use of a phage cocktail) that has an ecologically and evolutionarily-sensible Plan B (adaptively trained bacteriophage) should Plan A be partially successful or fail.

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Table 2. Opportunities and potential risks in treating disease associated with each of the 6 wedges.

Wedge	Opportunities	Potential risks *(Comments)*
1. Dosing	Hit hard, fast and sufficiently long if probabilities of sensitive DCA elimination are high; resistance low; immune system completes clearance	Toxicity and selection for resistance *(Modulate dose and schedule or manage using one or more of Wedges 2-6)*
2. Combinations	Break through refuges (phenotypic, dormancy, spatial, resistance); not only reduces DCAs compared to any drug used separately, but also may control drug resistance; combinations can be pre-evaluated for their synergy and used at lower doses than either drug separately	The development of multiple resistances *(Choose agents, doses and schedules whereby one agent is the principal treatment and the other acts as a supplement or adjuvant - Wedge 4)*
3. Increasing resistance costs	Targets the cause of many chemotherapeutic failures by reducing or eliminating resistant phenotypes before a therapy commences	Pre-therapy clinical side effects or causing significantly delays in administration of the drug *(Alternate with chemotherapies - Wedges 2, 6)*
4. Dynamic agents	Particularly useful as targeted therapies, to grow and persist in disease ecosystem, and counter evolved DCA resistance	Host range expansion to commensal microorganisms (bacteriophage); removal by the immune system; more time required for success *(Combine with more traditional drugs - Wedge 2 - or natural components of disease ecosystem - Wedge 5)*
5. Harnessing immune responses and quenching resources	Harness immune system, competition (public good quenching, nutrient limitation), and altering environmental stresses (reduced inflammation)	Autoimmune reactions (also risks in Wedge *4*) *(Most promising if used in combination with other therapies - Wedge 2)*
6. Adapting	Potential when the DCA can be directly or indirectly monitored and treatments be modified in type or dose, or adapted in parallel (e.g., phage training)	Inability to manage or eradicate infection

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The Greater Community

The approaches presented above focus on the disease ecosystem within the individual patient. For infectious diseases, one must also consider the larger population for at least two reasons. First, in the short term, failure to sufficiently control an infection may result in the transmission of sensitive, or even worse, resistant strains. Second, in the longer term, the continued use of a single antimicrobial over a sufficiently large population will invariably select for the emergence and spread of resistance. Heesterbeek and colleagues [74] presented a framework that reveals the complexity of managing infectious disease systems, whereby models (based on predictive epidemiological parameters, such as the basic reproductive rate), data and policy are integrated into a plan of action, and improved iteratively. These models have the objective of control (whilst preventing or delaying resistance) or, if possible, eradication. Application of one or more of the wedges proposed here should form part of disease control and eradication policies since, by reducing the probability of resistance emergence, they may prevent or attenuate the spread of epidemics in the wider population. Nevertheless, more research is needed to understand how attempting to cure individual patients is or is not optimal for the population, and inversely, how optimal population-level programs may result in some patients not being cured, whereas they would have been in a patient-centered approached. The 6 wedges presented here, when used singly or in combination may provide a way forward to optimize both individual outcomes and population-level outcomes.

Conclusions

The complexity of the disease ecosystem provides numerous opportunities for the employment of novel therapeutic approaches. Real disease ecosystems and the mechanisms of DCA escape are evidently much more complex than presented here (see e.g., Table 2 in [75]), meaning that compromises are necessary between sufficient realism and conceptual and computational tractability. This will be particularly challenging in situations where rapid decisions are needed and for which accurate, rich information about the DCA is difficult to obtain or difficult to process [76]. Nevertheless, the six wedges proposed here, based on a fundamental understanding of ecology and evolution in the disease ecosystem, provide a robust armory, which can be used singly or in combination to cure disease.

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