Rethinking Causation in Cancer through Modularity

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Abstract: Despite the productivity of basic cancer research, cancer continues to be a health burden to society because this research has not yielded corresponding clinical applications. Many proposed solutions to this dilemma have revolved around implementing organizational and policy changes related to cancer research. Here I argue for a different solution: a new conceptualization of causation in cancer. Neither the standard molecular biomarker approaches nor evolutionary medicine approaches to cancer fully capture its complex causal dynamics, even when considered jointly. These approaches map on to Ernst Mayr's proximate-ultimate distinction, which is an inadequate conceptualization of causation in biological systems and makes it difficult to connect developmental and evolutionary viewpoints. I conceptualize causation in cancer through the notion of modularity as a bridge between molecular biomarker approaches and evolution medicine approaches. A modularity-based approach requires the consideration of relationships between multiple levels of organization and the incorporation of different time scales, thereby overcoming the proximate-ultimate divide. The proposed perspective on causation in cancer is better suited to integrating the complexity of current empirical results and can facilitate novel developments in the investigation and clinical treatment of cancer.

Keywords: cancer, causation, evolutionary medicine, modularity, proximate-ultimate, biomarker

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1. Introduction

Numerous reports of cancer-related research are published each week, yet society continues to be underwhelmed by the rate at which discoveries are translated into effective clinical applications. In other words, cancer research appears to be progressing, but few new treatments or diagnostic and preventive tools have significantly impacted how doctors work clinically. Various parties have pointed to different reasons for this situation, including funding and institutional barriers. However, the translation failure rates remained high despite the doubling of the United States National Institutes of Health (NIH) budget between 1998 and 2003 (Sarewitz 2013). The NIH, the Canadian Institutes of Health Research (CIHR), and other funding organizations have recently emphasized translational research, an endeavor specifically focused on translating the knowledge gained at the bench into clinical applications. Much of the attention in translational research has been devoted to organizational and policy changes that remove barriers to clinical trials, redesign physician-scientist (MD/PhD) programs, and facilitate programs that encourage the formation of academic centers for interdisciplinary clinical and translational research (e.g., the Clinical Translational Science Awards consortium through the NIH). Much less attention has been devoted to whether the core research strategies used by translational researchers are adequate for the generation of desired clinical applications. It is assumed that the prevailing approaches and frameworks will translate into the clinic eventually, given enough time and the right combination of researchers. As I will argue, this is a bad assumption and therefore attending to institutional structure and research organization alone will be insufficient. Cancer translational research needs a more radical solution.

I am not the first to recognize this, nor the first to suggest solutions to this problem. Evolutionary and ecological approaches to cancer research emerged in response to the lack of clinical applications from the basic biology discoveries (Wodarz 2006; Greaves and Maley 2012; see more below), and the parallels between cancers and ecosystems have continued to be

elucidated (Merlo et al 2006; Kareva 2015). Physicists and mathematicians were recruited to ask questions about cancer in new ways, including using their mathematical and computational skills to model and investigate virtual cancer systems (Michor et al 2011; Agus and Michor 2012; Kuhn and Nagahara 2013). Philosophers also have sought to explain and make sense of cancers in novel ways (Bertolaso 2016), such as through multilevel selection (Lean and Plutynski 2015) or cancer stem cell theory (Laplane 2016). Although these suggestions have led to valuable research programs, they miss the key issue at the root of the difficulties facing cancer translational research: the conceptualization of causation in cancer.

The central thesis of this paper is that cancer translational research needs a new perspective on causation. In the next section (Section 2), I review two predominant approaches to investigating cancer (the molecular biomarker approach and the evolutionary medicine approach), as well as dissect their causal frameworks. Then in Section 3, I argue that the frameworks are insufficient because they map on to Ernst Mayr's proximate-ultimate distinction, which is an inadequate conceptualization of cancer's causation in biological systems. In Section 4, I suggest that this problem can be solved through a notion of modularity, by drawing an analogy to evolutionary developmental biology. I apply modularity directly to cancer translational research in Section 5.

2. Predominant Approaches and Frameworks for Investigating Cancer

2.1. Molecular biomarker approach

Biomedical research has become increasingly molecular with the availability of technologies such as whole genome sequencing. The promise has been that a better understanding of molecular mechanisms will make targeted clinical interventions possible. These interventions modify or correct abnormalities in particular individuals while minimizing side effects that come with traditional blanket treatments such as chemotherapy. The focus on

targeted interventions in individuals has fueled the rise of precision[1] medicine (Collins and Varmus 2015). One version that has become widespread is based on sequencing the genomes of individuals. The goals for this endeavor are to identify and develop drugs that will be effective for the patient based on their specific genomic composition. Because genomes can be sequenced, scanned, and compared quickly and (relatively) inexpensively, researchers look for variants that are biological markers (biomarkers) of diseases. These variants are supposed to then be differences between healthy and sick individuals that give insight into how patients will react to drugs, which allows for tailored prescriptions and dosages.

One of the most successful examples of the molecular biomarker approach is trastuzumab, also known as Herceptin (Genentech).[2] The FDA approved trastuzumab for the treatment of metastatic breast cancer in 1998 (Ignatiadis et al 2009), and it has been heralded as having a profound impact on the care of HER2+ breast cancers[3] (Hudis 2007), and as "a major advance in targeted cancer therapies" (National Breast Cancer Coalition 2013). Trastuzumab is a humanized monoclonal antibody with two binding sites specific to the extracellular portion of HER2, a cell surface protein associated with the formation of homodimers as part of a cell growth signal pathway. When HER2 is overexpressed, the homodimers continually signal cells to grow and divide. Trastuzumab binds to the HER2 protein and blocks dimerization, which prevents further activation of signaling. Additionally, the antibody triggers the immune system to target HER2+ cells with increased antibody-dependent cell-mediated cytotoxicity (Hudis 2007).

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[1] Though precision medicine and personalized medicine are different endeavors, there is much overlap between the two and the term 'precision medicine' is replacing the use of 'personalized medicine' (Katsnelson 2013).

[2] Imatinib (Gleevec), used to treat chronic myeloid leukemia (CML), is by far the most successful example of a targeted cancer therapy. However, CML is caused by a single chromosomal rearrangement, which means its causal dynamics are not widely applicable to other cancers.

[3] Breast cancers are classified as HER2+ when there is an overexpression of the membrane-bound HER2 (Human Epidermal Growth Factor Receptor type 2) protein. HER2+ breast cancers comprise 20-30% of invasive breast carcinomas, and these patients have decreased overall survival (Hudis 2007).

This example shows how the molecular biomarker framework can work. Researchers identified a mechanism (a cell growth signaling pathway), which is causally relevant to carcinogenesis, and a specific biomarker (the HER2 protein) where a targeted intervention should substantially make a difference. Original clinical trials with HER2+ metastatic breast cancer patients showed positive results in 12-25% of the cases, depending on previous treatment regimes, thus showing a proof of principle for the effectiveness of trastuzumab. In later trials, when administered in an adjuvant setting (i.e., trastuzumab plus chemotherapy), the risk of recurrence dropped by 40-50% and the risk of death dropped by one-third (Ignatiadis et al 2009). In comparison to most cancer treatments, these results are impressive. But, given the straightforward nature of the intervention, greater response rates were expected. Additionally, further analyses suggest that the chemotherapy accounts for much of the efficacy (Moja et al 2012). Some anticipate that identifying more tumor characteristics (including more biomarkers) will lead to better efficacy (Ignatiadis et al 2009). Others have suggested that the limited efficacy is due to misplaced attention on HER2 expression. That is, HER2 expression is a bad proxy. Karamouzis and colleagues (2007) suggested that heregulin, a ligand that binds to HER2 (and other) dimers, might be a better biomarker than HER2 overexpression.

The fact that trastuzumab was developed to treat invasive or metastatic cancer but shown to be effective only in early stage cancers[4] says something significant about the causality of cancer. The molecular biomarker approach might work if its target was part of a relatively static system, but diseases like cancer are dynamic and progressive. Molecular biomarker approaches ignore the developmental nature of cancer in individuals, and of disease biology more generally. Disease-affected systems in individuals are complex and have evolved robust and redundant pathways in order to address circumstances of disturbance. To investigate and

[4] Trastuzumab is still used to treat metastatic HER2+ breast cancers but in combination with other therapies targeted at stopping HER2 from forming dimers with other ligands (Baselga et al 2012).

understand these complex systems, many multilevel and integrated approaches are needed to account for causal dynamics involving feedback loops and robustness at different levels and across different time scales (Mitchell 2009). The molecular biomarker approach alone is insufficient.

Molecular biomarkers as difference makers

In order to see why the molecular biomarker approach is insufficient, it is important to look at the causal framework employed. Recall that this approach involves finding the molecular differences between healthy and unhealthy individuals or tissues and then intervening on those differences to change the resulting phenotype. Thus, the molecular biomarker is what makes the difference between being healthy and unhealthy.

Many experiments designed to understand causal structures are modeled on methods following John Stuart Mill's methods of difference: if you have two identical systems differing in only one factor, then differences in outcome can be attributed to that differing factor. Under these conditions, the factor is a difference maker. Knockout experiments are a widely used form of this causal reasoning strategy. For example, to infer the role of the gene *TP53* in cancer, one can compare standardized mice, some of which have *TP53* inactivated and some with functioning *TP53.* If mice with inactivated *TP53* show abnormal cell development that results in tumors, whereas the mice with functioning *TP53* show normal cell development, then, assuming all other factors are equal[5], we can infer that the *TP53* gene plays a causal role in cell development (Mitchell 2009). James Woodward's interventionist account of causation (Woodward 2010) is a more formal description of the causal framework of difference-making relevant to the molecular biomarker approach:

[5] This is the reason to use standardized mice. These mice are inbred to be genetically identical and raised in the same environments. This underwrites the assumption is that everything is identical, except for the knocked-out gene.

> *X* causes *Y* if and only if there are background circumstances *B* such that if some (single) intervention that changes the value of *X* (and no other variable) were to occur in *B*, then *Y* or the probability distribution of *Y* would change.

Using standardized mice in knockout experiments is a way to achieve the same background circumstances (*B*). The variable *X* takes two values: functioning *TP53* and inactivated *TP53*. The variable *Y* takes on two values as well: normal cell development and abnormal cell development. This gives the following causal formulation: if one intervenes on *TP53* and changes its value from functioning to inactivated, and the value of the normal cell development changes to abnormal, then one can infer that *TP53* causes (i.e., make a difference to) normal cell development. An important part of this formulation is the attention put on a particular place where a specific intervention can be made by the researcher to change the outcome of the causal processes, assuming that *B* remains fixed.

Despite the value of this causal framework, there are several limiting assumptions related to its application to different areas of medicine. The first assumption is that there is a one-to-one relationship between the variant and the disease phenotype; i.e., '*X* causes *Y*', not '*X* and *Z* cause *Y*' or '*X* causes *Y* and *W*'. Many-to-one relationships ('*X* and *Z* cause *Y*'), either as multiple pathways resulting in the same phenotype or many causal factors contributing to one phenotype, can make it difficult to know where to intervene because these cases facilitate redundancy in the system that, in some cases, compensates for the change. Additionally, interventions on one-to-many or many-to-many relationships ('*X* causes *Y* and *W*' or '*X* and *Z* cause *Y* and *W*'), such as genes showing pleiotropic effects in two or more seemingly unrelated pathways or phenotypes, can lead to unwanted outcomes. In other words, it is difficult to change *X* without affecting other variables. Thus, redundant pathways and pleiotropy will constrain the success of the difference-making causal framework used in the molecular biomarker approach.

A second assumption is that the background conditions remain the same when you change the value of the variable (e.g., from functioning *TP53* to inactivated *TP53*). That is, to test if functioning *TP53* makes a difference on normal cell development, one needs a group of individuals with functioning *TP53* and a group of individuals with inactivated *TP53*. Additionally, for this causal framework to apply, the activation of *TP53* has to be the only difference. This might be possible (or close to possible) in laboratory conditions with the use of standardized model organisms or cell cultures, but in the clinical setting this assumption does not hold. The same mutation can be malignant in one individual, but not in another depending on the genomic background or previous mutations (Greaves and Maley 2012). The variation from one individual to another often violates the condition that the background conditions are similar, and the partial effectiveness of Trastuzumab in treating breast cancer illustrates this clearly.

Finally, the difference-making framework tends to treat all difference-making factors symmetrically. That is, it is assumed that all difference-makers in that system will affect the outcome in similar ways, and there is no principled way to choose which factor(s) to privilege. However, not all causal factors play symmetrical roles in biological systems. Woodward (2010) suggests that knowledge of other characteristics of causal relationships, such as stability and specificity, will lead to more nuanced pictures of different causal relationships, though the conditions for stability and specificity are less precisely characterized than those for difference-making. Interventions relevant to the clinical treatment of cancer (e.g., via surgery or chemotherapy) require not only that a factor be identified as an actual difference-maker (versus a potential difference-maker), but also detailed knowledge of *how* it makes a difference (e.g., stability and specificity). Thus, identifying actual difference-makers permits the identification of multiple causes relevant to tumor progression or other aspects of cancer (Waters 2007), but does not, on its own, suggest further characteristics that would facilitate clinical applications.

It is important to note that cancer researchers may or may not think that these three assumptions–there are one-to-one causal relationships, background conditions are sufficiently

similar, and identifying difference-makers alone is adequate–are appropriate or true of the world. The point is that they are assumptions of the causal framework used to identify molecular biomarkers. Regardless of whether all of the researchers share each of the assumptions of the causal framework, they are embedded in the molecular biomarker approach. These methodological assumptions help to explain why this approach is insufficient to dissect cancer's complex causal dynamics.

2.2 Evolutionary medicine approach

Towards the end of the 20th century, a group of clinicians joined forces with a group of evolutionary biologists to reconnect medicine and evolutionary biology. Although a driving aspect of this enterprise was incorporating evolutionary biology into medical school curricula (Williams and Nesse 1991; Nesse and Schiffman 2003; Nesse 2008; Nesse et al 2010), the main research goals were to derive new insights into both medical and evolutionary questions, as well as change clinical practices (Williams and Nesse 1991; Nesse 2001; Nesse and Stearns 2008; Valles 2012; Stearns 2012). For example, can an evolutionary approach answer why particular symptoms appear? Why are certain individuals more susceptible to certain diseases (Nesse 2001; Nesse et al 2006; Nesse and Stearns 2008; Nesse et al 2010)? How should answers to these questions modify patient care?

Evolutionary medicine has answered many medical questions utilizing population genetic theory and an adaptationist research program (Nesse and Stearns 2008; Nesse 2008; Valles 2012). An important example is explaining the prevalence of sickle cell anemia through heterozygote advantage. Individuals that are homozygous for the recessive sickle-cell allele (SS) suffer from sickle-cell anemia, which is usually fatal due to the formation of blood clots and decreased oxygen delivery to tissues. Individuals that are homozygous for the dominant allele (AA) have normal red blood cells but are susceptible to malaria. Those that are heterozygous (AS) have both normal and sickled red blood cells, and have increased resistance to malaria, as

the sickled phenotype makes the red blood cells less suitable for the malaria-causing parasite. In environments where malaria is epidemic (e.g., many regions of Africa), the AS genotype is more fit than either the AA or the SS genotypes. Therefore, under these environmental conditions, the S allele will be maintained in the population and sickle-cell anemia will continue to persist. Note that this explains the persistence of sickle cell anemia but does not suggest any changes to clinical practices.

Antibiotic resistance, on the other hand, is common example used to demonstrate medically-relevant evolution in real time (Stearns 2012) and also has potential for clinical application. Resistance was long thought to be a result of evolved coexistence with the host. But selection leads to higher virulence of pathogens, in part due to rapid parasite evolution (Williams and Nesse 1991; Nesse and Stearns 2008; Nesse 2008), and thus clinical efforts to minimize resistance were misinformed previously. By administering large doses of antibiotics to reduce the chance of de novo resistance mutations occurring, clinicians were actually selecting for rare resistance genes already present in the population, which could then be horizontally transferred to otherwise susceptible bacteria. This has led to more discussions about the use of antibiotics in hospitals and clinics, including how to balance the killing of bacteria of an infection without strongly selecting for existing drug-resistant bacteria (Smith et al 2014).

A key contrast between the molecular biomarker approach and the evolutionary medicine approach is that the former concerns individuals and the latter populations. However, the examples of sickle-cell anemia and antibiotic resistance show that the appropriate level at which to consider “the population” in evolutionary medicine can vary from populations of people (e.g., in the sickle-cell anemia case) to populations of cells (or populations of microbes in the antibiotic resistance case). With respect to cancer, it is no different. For populations of people, mismatches between past and current environmental contexts can explain the persistence of vulnerabilities to cancer (Greaves 2002; Aktipis and Nesse 2013). Additionally, population genetics can be applied to cancer incidence data to test hypotheses about the causal roles of

mutation and drift in cancer (Frank 2007). This informs predictions about when incidence curves will shift, and thus when to start screening for particular cancers in certain populations.

With respect to populations of cells, Peter Nowell proposed a model of clonal evolution in tumors in which they develop over time through a series of stepwise somatic mutations followed by repeated selection (Nowell 1976). A mutation occurs in a somatic cell (either spontaneously or induced by a carcinogen) that gives it a growth advantage over neighboring cells and facilitates the clonal expansion of cells containing that mutation. As the population of cells expands, more mutations accumulate, some of which will give additional selective advantages, allowing those subpopulations with the advantage to be selected for and further expand. This process results in tumors that reflect specific and local environmental pressures. A more nuanced account of this process treats the tumor as a byproduct of the tissue ecosystem (Greaves and Maley 2012). Each time a treatment (e.g., chemotherapy) is given, it changes the ecosystem, which changes the development of the tumor. As a result, if part of the tumor is removed, then the tumor that recurs is a different tumor than what was originally there. This model illuminates why there is variation within a tumor and differences between individuals with the same kind of cancer.

A primary goal of evolutionary medicine is to have clinical impact. By analyzing incidence data from populations of people, we can develop screening policies. By applying evolutionary theory to populations of cells, new treatment regimens are suggested, such as adaptive therapy, where the goal is to maintain tumors at a stable size rather than fully eliminate all cancer cells (Gatenby et al 2009; Enriquez-Navas et al 2016). Cytotoxic drugs select for resistant cells and create an environment where the resistant cells can proliferate, causing a tumor to become more aggressive and harder to treat. As with antibiotic resistance, investigating cancer through an evolutionary lens can lead to better uses of existing treatments. However, this approach yields little mechanistic information that is relevant for the development of new treatments. More poignantly, an evolutionary framework does not help treat metastasis,

the pathological form of cancer that is responsible for most deaths. The fact that an evolutionary medicine approach gives predictive power and thus offers preventive possibilities is incredibly important. However, not every case of cancer can be diagnosed early, nor should we necessarily depend on early detection and prevention (Narod et al 2015; Prasad et al 2015). Thus, we still lack effective approaches for the investigation and clinical treatment of cancer, especially metastatic cases. The evolutionary medicine approach alone is insufficient.

Natural selection and population-level causes

Like with the molecular biomarker approach, it is useful to look at the causal framework employed by an evolutionary medicine approach. Population genetics is the basis of evolutionary medicine (Nesse and Stearns 2008; Nesse 2008), which means natural selection, mutation, drift, and migration are factors in its causal framework. However, in practice, evolutionary medicine prioritizes natural selection as a cause, making it an adaptationist program (Valles 2012). For example, its answer to why we are still vulnerable to certain diseases is that humans are "bundles of compromises shaped by natural selection … to maximize reproduction, not health" (Nesse and Stearns 2008, 28). That is, much of the research program focuses on maladaptations (Nesse 2005). Individual organisms in a population show heritable variation with fitness differences, but natural selection is a population-level cause (see, e.g., Millstein 2006). Consequently, the causal framework of evolutionary medicine does not speak to the condition of specific individuals in the population.

Notably, mutation, drift, and migration play more prominent causal roles in evolutionary approaches to cancer than in conventional evolutionary medicine (Merlo et al 2006; Aktipis and Nesse 2013). Mutations create the heterogeneity in a tumor allowing for faster progression and therapeutic resistance (Aktipis and Nesse 2013). Genetic drift occurs when population sizes are small. While tumors are generally large populations of cells, metastasis (the spread of cancer to other locations in the body) occurs via small clusters of cells which create population

bottlenecks (Aceto et al 2014). Additionally, cancer stem cell populations are usually composed of only a few cells (Baker et al 2014), which explains why deleterious mutations can go to fixation and why some tumors might be more aggressive than others. Migration plays several roles in cancer biology. One is related to mismatches between ancestral conditions and modern environments. For example, light-skinned individuals migrating towards the equator creates a mismatch between skin pigment and sun exposure, and thus changes their susceptibility to skin cancer (Aktipis and Nesse 2013). Migration might also be important in explaining metastasis, which inherently involves the migration of cells from one location to another. One model suggests that heterogeneity in a tumor selects for migratory cells (Chen et al 2011). Although these evolutionary insights into cancer complement what has been discovered in terms of molecular mechanisms, they have not yet had a clinical impact. They account for why it is important to catch and treat cancers early, but do not give mechanistic information that would be useful for treating existing cancers in specific individuals. As a consequence, evolutionary medicine approaches alone are insufficient to dissecting cancer's complex causal dynamics.

3. The problem of the proximate/ultimate distinction

Combining the molecular biomarker and evolutionary medicine approaches?

Although both the molecular biomarker and evolutionary medicine approaches are necessary, neither is sufficient to dissect the causally complex dynamics of cancer, even when considered jointly. The approaches ask fundamentally different questions and therefore investigate causes using different frameworks. Researchers using the molecular biomarker approach want to know *how* cancers arise and progress in individuals, and *how* specific molecules make a difference to the outcome or phenotype. Alternatively, those using the evolutionary medicine approach want to know *why* cancers occur and progress, and *why* certain

populations are more susceptible. The causal framework differences between the approaches fall on opposite sides of Ernst Mayr's proximate-ultimate distinction.

Mayr distinguished between two different kinds of causes: proximate and ultimate (Mayr 1961). Proximate causes are those studied by functional biologists (e.g., physiologists and developmental biologists) and often answer "How?" questions. Explanations that come from this orientation involve immediate factors, such as day length or hormone levels. For example, a change in hormone levels causes an organism to change its behavior. In contrast, ultimate causes are those that involve evolutionary history and are present due to many generations of natural selection. These explanations are historical in nature and often answer "Why?" questions. For example, an organism has a behavior because it was beneficial to its ancestors and was selected for within a population over many generations. Mayr aimed to differentiate proximate and ultimate causes while arguing that both kinds of causes are necessary for a complete understanding of any biological phenomenon. Thus, according to Mayr, the molecular biomarker and evolutionary medicine approaches might together yield a complete causal framework for understanding causation in cancer. Although having both approaches does give a fuller causal picture than just one approach alone, there are reasons why we might not want to interpret the biomarker and evolutionary medicine approaches as separate or distinct in this way.

Kevin Laland and colleagues have argued that the distinction between proximate and ultimate causes, though helpful at times (e.g., it prevents scientists from talking past each other, allowed evolutionary biology and developmental biology to independently grow and mature as disciplines, etc.), has actually hindered progress in biology, particularly evolutionary biology (Laland et al 2011; Laland et al 2012). This is because the distinction has become entrenched as a truth about the nature of causation, rather than as a heuristic in the study of causes (Laland et al 2012); it has shifted from being a good way to approach causality in biological systems to being the only way one can understand causation in biological systems. The mentality has

become entrenched because Mayr's basic premise–proximate and ultimate causes answer different questions and are not in competition with one another–implies that there is no reason to link them; proximate causation is not important to the investigation of ultimate causes and *vice versa* (Laland et al 2012). This makes it difficult for the approaches to mutually inform one another in a way that would contribute to clinically relevant outcomes. This mentality is manifested in cancer research, where the two approaches are funded through different avenues and the research communities rarely overlap.

Maintaining the proximate-ultimate distinction leads to other problems as well. First, Laland and colleagues argue that this distinction cannot account for cases of reciprocal causation (Laland et al 2011; Laland et al 2012). If developmental processes are interpreted as proximate causes and separated from evolutionary processes interpreted as ultimate causes, we miss the fact that developmental processes contribute to the phenotypes on which selection will act, and these mechanisms are under selection themselves. Thus, developmental processes (proximate causes) are both an input and an output of ultimate causes (e.g., selection). Second, the proximate-ultimate distinction and how-why distinction are false dichotomies because it is possible to ask questions that can be answered with both or neither category (Calcott 2013). More generally, it is not clear what it means for an explanation to be historical, which is presumed in the proximate-ultimate distinction. This affects the range of possible answers to ultimate questions and indicates that we are getting only partial answers or explanations.

While these are legitimate concerns, these and other critiques of the proximate-ultimate distinction arise mostly from those working on evolutionary questions. Evolutionary biologists are concerned about whether evolutionary theory needs to be extended to include developmental biology and other causes typically categorized in proximate causation (Laland et al 2014). But little to no critique of the distinction has come from developmental biologists. Do studies typically categorized as proximate causation (such as developmental biology) require

studies typically categorized as ultimate causation (such as evolutionary biology)? This is a more applicable question in the context of cancer translational research.

The molecular biomarker approach is predominant (by the amount of money allocated, number of research programs, papers, etc) in the methodology of developmental biology. Although discovering molecular mechanisms is essential for knowing where to intervene on individuals with cancer, an evolutionary perspective informs when to intervene on a population. But simply identifying or acknowledging the proximate-ultimate distinction does not facilitate bridging the causal frameworks of the two approaches. Each approach is complementary to the other, but just having both side-by-side is not enough. That is, there are probably important factors or dynamics that are being missed (e.g., reciprocal causation). The standard molecular biomarker and evolutionary medicine approaches do not fully capture the complex causal structure, even when considered jointly. We need a perspective on causation that integrates both developmental and evolutionary approaches to cancer in order to better understand cancer and identify effective clinical treatments.

4. Bridging the proximate-ultimate divide through modularity

Bridging the proximate-ultimate divide is not a trivial task; proximate and ultimate causes are typically studied using different methodologies, work on vastly different time scales, and depend on different underlying conceptual foundations. Similar to cancer translational research, evolutionary developmental biology (evo devo) aims to explore the connections between the development of individual organisms and their evolutionary transformation to discover causal-mechanistic explanations of individual traits involved in population-level events (Laubichler 2007; Hamilton 2009). One way evo devo has pursued these connections is through the notion of modularity (von Dassow and Munro 1999). By analogy, I suggest that cancer research can benefit from looking to modularity as a way to reconceptualize causation in cancer.

Modules and modularity

Although there are many definitions of modularity, a module is generally considered to be an entity or part that is discrete or *autonomous* in some ways but *integrated* within a larger whole (usually the rest of the organism) in other ways (Wagner et al 2007). Modules can be processes or structures (Raff 1996), and be instantiated at any hierarchical level. Examples include: fragments of *cis*-regulatory DNA (Arnone and Davidson 1997), morphogenic fields, signal transduction pathways, gene regulatory networks (Gilbert and Bolker 2001), and leaf primordia (Gass and Bolker 2003). Furthermore, organisms themselves can be considered modules of higher-level individuals, such as superorganisms (Schlosser and Wagner 2004). Thus, modules can be embedded in higher-order modules (Schlosser 2004).

Modularity involves part-whole relationships, but various fields think about these relationships differently. Evolutionary biologists consider modules to be dissociable subunits or parts of a larger system, typically the adult organism (Gass and Bolker 2003). Evolutionary modules are parts autonomous enough to change without appreciably changing other aspects of the organism. Rasmus Winther (2005) calls this a partitioning strategy, where you can understand the whole as a sum of the parts. On the other hand, developmental biologists refer to modules as collectives of entities and processes that act in some unified way to perform a function (Bolker 2000). In this view, a strategy of articulation is used when the relationship or interactions between parts is just as important, if not more, than the parts themselves (Winther 2005). For evolutionary approaches, the autonomy of a module is usually foregrounded, whereas in developmental approaches, the interactivity or integration between modules is more important.

Evo devo combines these approaches to modules so that evolutionary modules are the phenotypes that result from particular developmental modules (Gass and Bolker 2003). However, there is not a one-to-one relationship between developmental modules and resulting evolutionary modules (Bolker 2000; Schlosser and Wagner 2004); multiple modules of the same

type can be used in a single pathway or process, or modules can overlap by sharing elements (Schlosser 2004). Thus, what is critical for evo devo researchers in bridging developmental and evolutionary approaches is how modules are individuated and interact with one another. Once meaningful modules are identified, the interactions between them become the focus because it is the *changes in interactions* that result in phenotypic differences and therefore evolutionary change[6].

Using modularity as a way to bridge different causal frameworks forces us to shift our attention in two main ways. First, as was discussed above, we shift our attention away from investigative strategies that are specific to proximate or ultimate causation, and thereby avoid treating proximate and ultimate causation separately. Second, as I will discuss below, a causal perspective based on modularity inherently requires the consideration of relationships between multiple levels of organization and across different time scales.

5. Cancer translational research and the causal framework of modularity

It is important to emphasize that my argument is not that a modularity-based causal perspective can or should bridge the proximate-ultimate distinction across biology generally, but rather that it can bridge the proximate and ultimate causal frameworks in cancer translational research specifically. As noted, there are multiple ways to conceive of modularity–by focusing on the autonomy of the modules, the interactivity, or the changes in interactions. Since cancer is both a developmental and evolutionary phenomenon, I continue with evo devo's approach to modularity–modularity can occupy the spaces of both proximate and ultimate frameworks, thus bridging the divide and highlighting the causal complexity of cancer.

[6] The agnathan-to-gnathostome transition is a good example of this, where heterotopic (spatial) shifts in developmental modules resulted in the gnathostome-type oral apparatus in which both the upper and lower jaws are derived from the mandibular arch. In contrast, only the lower lip of the agnathan is derived from the mandibular arch. The upper lip is derived from the premandibular module (Kuratani 2009).

Evo devo's interpretation of modularity was not meant to replace the evolutionary or developmental interpretations with an intermediate notion, but rather allow for the discussion of modularity at multiple levels and across time scales which requires being clear about which notion of modularity is being used for investigation. For example, one can have discussions about modularity at the level of individuals or populations. Thus, having a causal perspective based in modularity requires first identifying meaningful modules for investigation. There are many potentially meaningful modules in cancer – molecular pathways, cells, tumors, tissues, the circulatory system, the immune system, and many more. By conceptualizing causation in cancer through modularity, the changes in the interactions between identified modules as the cancer progresses (or regresses), take on a new significance. Cells normally interact in specific ways to form tissues, but when a cell gains enough autonomy (i.e., it becomes highly modular) to defect from or cheat within the tissue, there is the possibility for it to become cancerous. As the cell grows into a tumor, it progressively loses some autonomy and becomes integrated with other cells in the tumor. As the tumor continues to grow such that nutrient and waste transport become important, its dependence on angiogenesis (the development of new blood vessels) and interactivity with the circulatory system change. Additionally, when treatment is administered (especially immunotherapy), the cancer forms a different complex, iterative relationship with the immune system. The interactions between modules and the relationships between the parts and whole are constantly changing depending on the tumor microenvironment, as long as the cancer exists. These multilevel changes in relationships across different time scales facilitate and constrain the evolvability of the cancer in certain ways, thus affecting any possible cancer-related treatments.

Additionally, knowing which parts of the organism to consider as meaningful parts and wholes is only possible if cancer is considered with a temporal aspect. Different modules become important during different stages of cancer progression. During tumorigenesis, the meaningful part might be the cell and the meaningful whole might be the tissue in which that cell

resides. Later in tumor growth, the meaningful relationships to study might be between the tumor and the circulatory system. Furthermore, during metastasis (the spread of cancer to new locations in the body), it might be important to investigate the relationships between the clusters of cells that break off, the circulatory system through which they travel, the immune system from which they must evade, and the new tissues in which they ultimately plant.

Major transitions in the progression of the cancer (e.g., from localized tumor growth to metastasis) are especially important places to find meaningful changes in interactions that could be manipulated for interventions. Tracking modular, multilevel causal interactions through different time scales in the progression of cancer has the potential to open up novel clinical treatment options that were previously hidden in the divide between proximate and ultimate causation. For example, immunotherapy appears to bridge proximate and ultimate perspectives by striving for the precision of the biomarker approach but also taking advantage of the evolving immune system to combat evolving cancerous systems. Researchers want to find proteins that are over-expressed in all and only cancer cells (such as PD-L1; (Iwai et al 2002)) across all time points so that the immune system can continue to target and kill the cancer cells regardless of how the cancer evolves or its state of progression. However, if we apply what I have discussed here regarding the complexity of multilevel causal interactions across time, we can see there is still much more work to be done in order to predict and manage the side effects (e.g., autoimmune disorders) and potential complications or failures of immunotherapies (especially regarding metastasis and recurrent tumors).

Remember that modules can be embedded within other modules and modularity is found on a spectrum–any given module can be more or less modular than another module–and the degree of modularity can change given changes in interactions. Modules that are less modular will be affected more when interactions between modules change than modules that are more modular. Thus we can expect that that as a given cancer progresses, we have to shift

which modules get highlighted. But we cannot forget that the relevant modules are interacting with other modules, as is the worry with the development of autoimmune disorders.

Reconceptualizing the causation of cancer through modularity requires that developmental and evolutionary questions be asked and answered to know which modules are worthy of attention. For example, questions about evolvability follow directly from a modularity-based framework because modularity is one among many ways for core molecular processes to confer evolvability (Kirschner and Gerhart 1998). If evolutionary medicine approaches desire to make a clinical impact, studies need to be prospective as well as retrospective. Generally, understanding the molecular mechanisms that are both causes and consequences of changes in modularity will be informative for understanding how changes in modularity affect evolvability. This also applies to cancer translational research, where meaningful interpretations of identified molecular biomarkers of cancer along with the predictive work from evolutionary medicine can be translated into clinical applications.

6. Conclusion

The molecular biomarker and evolutionary medicine approaches to cancer translational research use widely accepted methods of investigation and explanation in biology and biomedical research. These approaches lend themselves well to the current scientific culture as they produce quick results for publications and grants, and ultimately give simple and clean explanations that are easy to understand and convey to others, including the general public. However, they only occasionally produce or lead to “successful” interventions. Additionally, these interventions only help a small percentage of the population (see the herceptin discussion above), or are quite far from clinical implementation (e.g., adaptive therapy). Therefore, following Mayr’s suggestion to have separate proximate and ultimate causal frameworks does not work for cancer translational research, nor is it necessarily better than having just proximate or just ultimate causal frameworks. This is because cancer translational research requires more

than explanation and understanding. For example, knowing that you were more susceptible to a certain kind of cancer because of an upregulated pathway that gives those cells a fitness advantage does not tell one where to intervene in order to prevent or eliminate the cancer. In other words, the evolutionary medicine population approach does not isolate difference makers. Furthermore, oncologists and society alike would prefer interventions that work without first almost killing the patient (in theory, enough radiation or chemotherapy would kill all of the cancer cells, but the number of healthy cells also killed is far greater than zero). If the goal is effective and relatively safe interventions, we need a framework that embraces the causal complexity of cancer biology and transforms proximate and ultimate investigative strategies into one more cohesive and fuller explanation.

In this paper, I proposed a novel perspective on causation in cancer through the notion of modularity. I argued that the standard research programs fall too neatly along the Mayr proximate-ultimate distinction, but that in the context of cancer translational research, this distinction can be bridged through the notion of modularity. The multiple notions of modularity play a translational role that allows for an expansion of the set of causes one can consider in cancer translational research, so long as one is clear about which notion is being employed. Similar to what has been done within evo devo, the notion of modularity allows cancer translational researchers to have discussions that consider both proximate and ultimate causes at the level of the individual or population. By understanding mechanisms through which the relationships between various modules (cell, tumor, tissue, immune system, etc) change over different scales of time, cancer translational researchers can better comprehend the complexity of the molecular data as well as use the evolvabilities of different cancers to proactively develop treatment regimes, rather than continue to treat patients reactively.

Both of the current predominant approaches suggest catching and treating cancers early is the best option. Though shown true in some cases, this is not always possible, nor always the best option (see, e.g., Narod et al. 2015; Prasad et al. 2015). Therefore, it is essential to

develop treatment regimens for advanced and metastatic cancers. The modularity-based causal perspective described here is one way forward for a research program in cancer translational research that does not deny the vast amounts of existing data, but provides a path to discover and interpret these results for more effective treatments and foster future investigations that identify novel clinical applications.

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