THE TWO SIDES OF THE HOURGLASS: ANALYTIC AND SYNTHETIC APPROACHES IN CANCER RESEARCH

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ABSTRACT. Consolidation of experimental science has brought about the triumph of the analytic perspective that decomposes nature in order to understand its molecular instances. This methodological approach reinforced the reductionism that has dominated empirical research in biomedicine over the last century. Cancer research constitutes an example. Nevertheless, the evolution of the interpretative models of its etiopathogenesis shows how different levels of biological organization might be involved in cancer origin and progression. New models have been challenging traditional reductionism, moving towards a systemic view that is posing an epistemological stance in cancer research, revealing the potentialities beyond a synthetic perspective in studying biological phenomena and showing how the level of causal explanation become crucial to understand cancer. A new reflection on the philosophy of causation seems to be required through the integration of both perspectives, in order to provide a comprehensive causal account of the neoplastic process.

KEY WORDS. Cancer research, reductionism, biological complexity, biological causes, prediction, control.

INTRODUCTION

Experimental science permitted unimaginable progress in the knowledge of biological phenomena over the last century, which has allowed humankind to dominate processes for the cure and prevention of many diseases. When facing the complexity of biological events, experimental research has to analyze the mechanisms involved in these processes in order to understand the organization, interaction and integration of the elements that make up their phenomenology. The question about their explanatory causes forced the focus to move up to different levels of biological organization and structure. The effort to reconstruct the complexity of the processes of the living beings has then become the main challenge in many areas of biomedical activity.

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The evolution of interpretative models of cancer reflects this path. The analytical approach, which tries to decompose a phenomenon into its constitutive parts, allowed the collection of a huge amount of data, which have enriched our knowledge of the neoplastic process. Reductionism, which dominated biomedical research in the last century, assumed the analytical perspective as the main explanatory perspective. The pursuit for causes at the lower level of biological systems, provided the initial descriptive models of tumor progression, where genes were identified as the main causes of cancer. However, a high degree of heterogeneity in the mechanisms at the molecular level is still present in those models of cancer, making our understanding of them clearly insufficient in accounting for the initiation and progression of cancer, limiting the efficacy of many current therapeutic approaches. As scientists explore the molecular basis of cancer biology, the complexity of this disease became more and more evident. Interrelations between biological parts and processes, and the dynamical aspect of the neoplastic process, underlined the need for a more comprehensive perspective to account for them. The concept of 'emergent properties' has been used to refer to the new properties which appear at higher levels of biological organization that cannot be explained just through the properties of the parts of the system they belong to: a synthetic perspective is required to explain them. A progressive integration of explanatory causes at higher levels of biological organization in the interpretative models of cancer is generating a different orientation within cancer research, driven by a new systemic outlook. New theories and models arise that identify critical biological events in cancer progression up to the tissue and organic level.

In this paper, we review these changes in research on cancer, which seems to be well described by the metaphor of an hourglass. The two sides represents the history of etiopathogenetic models of cancer, through the identification of its causes; while their evolution sheds light on the scientific contribution of different epistemological points of view on scientific activity. We explore some aspects of the nature of scientific explanation in cancer research, which seems to contribute to a new reflection about the philosophy of causation as well. Some reasons for the integration of the analytical and synthetic perspectives are discussed in order to direct a huge amount of knowledge into a deeper understanding of the neoplastic phenomena.

FROM THE ENVIRONMENT TO GENES

Defined essentially in terms of prognosis and of the neoplastic masses observable by pathological analysis, cancer and its etiology were studied scientifically starting from the nineteenth century. The evolution of these

investigations shows a movement from a vision of cancer as an environmental disease to the still present view that looks at cancer mainly in terms of a genetic disease. In 1863, Rudolf Virchow (1821-1902) published an article in which tumors were classified on the basis of their morphology, supporting the idea that cancer was related to both endogenous and exogenous factors as well as natural and social events. The first intuition that the pathology affected cells and their progeny also belongs to him. The idea that cancer might be a cellular pathology was confirmed later by microscopic observations, which revealed an elevated disorganization of chromatin in the tumor cells. This added a new level of structural and morphological disorganization to the characteristics of tumoral masses, which was confirmed by successive discoveries on DNA and the molecular basis of hereditary genetics.

Interestingly, although it was Virchow who first proposed that inflammation had an important role in tumor initiation, many years passed before its role became better understood and this organical level of analysis was again taken into account. During the latter part of the twentieth century, while interest for initial pathological studies was lost, cancer etiology remained at the forefront of the research efforts, although the attention was mainly focused on the physical and chemical aspects of carcinogenesis. The National Toxicology Program lists more than two hundred chemical, physical and infectious agents that are recognized as probable environmental carcinogens (National Toxicology Program 2005), yet carcinogenic processes are very specific, and the great majority of environmental chemicals are not known to be carcinogenic. The first physical cancer-causing agents identified included ionizing radiation and ultraviolet light. Even so, a major advance in cancer etiology was made with the study of chemical carcinogens. The chemical origin for human malignancies has been demonstrated through observations of unusual cancer incidences in certain occupational groups and population-based studies (Parkin 2004; Colditz, Sellers, et al. 2006). At a molecular level, although some chemicals that cause cancers in laboratory rodents are not demonstrably genotoxic, nevertheless both genotoxic and non-genotoxic carcinogens have been shown to alter gene expression through induction of DNA transcription or histone methylation, or other nuclear mechanisms that influence the transcriptome (Jones and Baylin 2007).

Even if DNA was identified as the target of carcinogens, it was not known, at the beginning, what genes were involved in the process. The study of the genetic basis of cancer is the cornerstone of modern cancer research, initiated when Boveri investigated the association between aberrant mitoses and malignant tumors (Boveri 1914) through experimental manipulations of sea urchin eggs by inducing multipolar mitoses and aberrant chromosome segregation. Unlimited growth—a common charac-

teristic of malignant tumors—was attributed to the incorrect combination of chromosomes, thus laying the foundations for viewing cancer as a genetic disease. The first genetic defect to be associated with cancer was a small chromosome identified in cancer cells of patients with chronic myelogenous leukemia (Nowell and Hungerford 1960). In 1980, David von Hansemann described the mitotic figures of thirteen different carcinoma samples, and all showed aberrant mitotic figures. Together, these ideas found a widespread acceptance as molecular explanations of cancer. Starting from the 1980s, new avenues of research were opened for the etiopathogenetic study of cancer with the discovery that certain viruses were able to induce tumors. Although several DNA viruses are associated with the development of malignancy, members of two RNA virus families have also been associated with development of neoplastic disease (Klein 2002; Burmeister 2001). After Rous's discovery that an avian tumor could be transplanted to other individuals, it seemed clear that a biological agent could directly cause tumors (Rous 1910). The agent discovered by Rous was subsequently shown to be a virus, and was named after its discoverer as Rous sarcoma virus (RSV). Later, it was demonstrated that RSV contained an oncogene that conferred tumorigenic properties in chickens. These studies had an enormous impact in the study of cancer as they definitively focused attention on the genetic component of the neoplastic growth.

While it is now known that only a few cancer types can be attributed to viruses, at that time many held the view that cancer was caused by infective agents, and the mystery of the cellular transformation process with its seemingly simple principles reaffirmed this idea. Research into the function of the protein products encoded by oncogenes followed closely after the discovery of oncogenes themselves, as in the case of platelet-derived growth factor (Waterfield 1983; Doolittle, Hunkapiller, et al. 1983). Successive empirical evidence, in fact, showed that some oncogenes found in retroviruses encode components of the normal growth-regulatory machinery of the cell, while others were shown to encode proteins that bind to kinases as modulators or signal transducers of specific cellular signals. Some researchers began exploring the biological meaning of the relationship between cancer genes and differentiation or development, although the bulk of the research community still viewed cancer as a fundamentally genetic disease, and the efforts of many researchers were directed towards the search for mutations that could explain the neoplastic process.

It was therefore proposed that mutations in cellular homologues of viral genes could transform cells in the absence of any viral involvement, and that such a phenomenon occurred in a substantial proportion of human cancers. Key discoveries showed that transformation could occur when the DNA of a chemically mutagenized transformed mouse cell was transferred to non-neoplastic cells (Shih, Shilo, et al. 1979; Cooper, Okenquist,

et al. 1980). However, the precise identity of the transforming gene remained unknown, and a large amount of potentially irrelevant DNA was also transferred in the process. Finally, in 1982, the groups of Weinberg and others cloned the first oncogene, from a bladder carcinoma line, after identifying the relevant DNA by numerous rounds of transfections. In each round, more of the transferred DNA was lost, until the oncogene could be cloned with the use of linked sequence tags. These cloned cellular genes had the same transforming properties as the oncogenes from retroviruses. Having uncovered the presence of cellular oncogenes, attention was turned to their identification. By the end of 1982, a single amino acid change, which altered the structure of the RAS protein to make it constitutively active, had been discovered and the first activating mutation had been identified. The developments in 1982 were a crucial step towards the modern understanding of cancer as a complex interplay between different types of genetic lesions (Shih and Weinberg 1982), and the role of oncogenes in cellular transformation became a main focus in cancer research. In the following years, many genetic studies conducted on the genetics of cancer and hereditary and predisposing factors provided hope for more efficacious strategies for prevention, diagnosis and therapy. The concept of mutation was thus introduced to explain the change in functional identity of a normal, somatic, cell into a tumoral cell, whereas traditionally the term mutation meant just a change in DNA sequence. The Somatic Mutation Theory (SMT) is based on this assumption.

The available evidence indicated that oncogenic transformation of primary cells involved at least two stages: establishment—also identified with immortalization of cells—and cellular transformation. With this in mind, several groups began investigating how oncogenes cooperate to induce tumor development (Land, Parada, et al. 1983; Ruley 1983) by looking at the effects of oncogene expression, assuming that these genes could display a dominant inheritance modality, like in the retinoblastoma syndrome. In contrast, the observation that normal mouse cells were dominant to malignant cells when the two types were fused (Harris 1971) suggested that cells had genes to oppose tumorigenesis, and some had a tumor suppressive function. This and other data (Harris, Rawlins, et al. 1996; Steel and Harris 1989) suggested that carcinogenesis does not require the acquisition of some new function, but rather the disruption of the pattern of cellular differentiation. This conceptually simple, though technically demanding work led to the theory that (dominant) oncogenes were not necessarily the general rule. Knudson postulated that predisposition could arise as the consequence of a heterozygous germline mutation in a tumor suppressor gene (first hit), while a subsequently acquired somatic mutation would be required for the tumor to develop (second hit). The model assumed that the second hit would take place in the remaining

normal allele of this hypothetical tumor suppressor, hence resolving the paradox (Comings 1973). Analysis of retinoblastoma, neurofibromatosis and childhood leukemia furthermore seemed to confirm this hypothesis (Knudson 1971). The two hit hypothesis found increasing support, but lacked additional insight into the nature of the hits.

Bert Vogelstein shed light on the role of oncogenes and tumor-suppressor genes by describing how alterations in both are necessary for colorectal carcinogenesis. In a pivotal paper, Fearon and Vogelstein presented these findings, together with the idea of clonal evolution, into a coherent molecular Multistep Model of tumorigenesis (Fearon and Vogelstein 1990). They therefore considered the total accumulation of changes, rather than their sequence, as most important for the tumor progression. They also concluded that five or more genetic alterations were probably required for the development of carcinomas, with even fewer changes needed for benign tumorigenesis. This Multistep Model was widely accepted by the scientific community (Vogelstein and Kinzler 2004), since it furnished the scientific basis for the initiation, promotion, transformation, and progression of cancer, which to that date had been only purely theoretical. The model also had the advantage of clarifying the genetic mechanisms in the role of initiation and progression of tumors. Moreover, it provided the promise for further studies on the neoplastic process in which the tissue and epigenetic components play a fundamental role. Researchers eventually defined carcinogenesis through errors in proliferation, cellular death or differentiation (Lloyd, Obermüller, et al. 1997), and adopted the working hypothesis that mutations are the causal agents of tumorigenesis and that neoplastic growth is a simple cellular phenomenon, even though it was still insufficient to explain tumorigenesis at large. We see then, that over the last century, the object of inquiry has moved from the environmental to the genetic level. In the following section, we will analyze how research is showing now an inverted orientation.

FROM GENES TO CELLS AND TO THE ENVIRONMENT AGAIN

Few would argue that the path to scientific discovery is short and simple. Initial evidence seemed to indicate that only a small number of processes were responsible for cancer, yet the number of factors and potential genetic causes has continued to grow. Given this, the functional role of all these elements still needs a clear and unified explanation, which seems to be in contrast to a mere reductionist view of cancer, that shows strong analogies with complex biological phenomena (Kitano 2005). Cancer could be seen not as an event, but as a multiphase process that depends not upon a single gene but from a cascade of events that lead to neoplastic transformation, involving different levels of biological organization. Mov-

ing from the SMT, several theories followed that attempted to provide a more unitary vision of the mechanisms underlying the neoplastic process.

When researchers began to take into consideration the role of epigenetics, the prospects for identifyng the causal factors for tumorigenesis and its progression, in addition to classic genetic factors, were greatly enlarged. Epigenetic phenomena can be defined as heritable changes in cellular information not contained within the DNA sequence itself, which usually involves covalent modifications to DNA or histones, and is often involved in the control of gene expression. Experiments in the 1980s (Jones and Taylor 1980; Feinberg and Vogelstein 1983) demonstrated reduced DNA methylation, an enzymatic reaction related to the regulation of specific genes, compared with DNA from adjacent normal cells, while more direct evidence linking DNA hypermethylation with cancer came several years later. At that time, changes in DNA methylation were believed to occur early and ubiquitously in cancer (Feinberg, Ohlsson, et al. 2006). In this regard, it was observed that the first mutation that led to Loss of Heterozygosity (LOH) could also be an epigenetic germ-line mutation, that could either substitute a somatic mutation or lead to an epigenetic bi-allelic inactivation. Epigenetic alterations, moreover, were demonstrated to be aberrations not to be found in just a single gene but in a genetic cluster, and led to the hypothesis that these modifications went beyond simple mutations as they could have an effective role in epigenetic control. The Epigenetic Progenitor Model (EPM) seemed to be able to explain the late onset of most adult cancers, environmental effects, tumor heterogeneity and to integrate the genetics of cancer risk.

Interestingly, the EPM created a new framework, integrating different hierarchical levels of biological complexity to the causal explanation for the first steps in the neoplastic process, opening up a new cellular level of explanation to cancer, through the concept of the Cancer Stem Cells (CSCs). A Hierarchical Model of cancer implies that only a small subpopulation of tumor stem cells can proliferate extensively and sustain the growth and progression of a neoplastic clone. The bulk of our understanding of CSCs has come from the study of hematopoietic malignancies (Furth and Kahn 1937). Those studies, along with others that followed, uncovered the functional heterogeneity present in tumors—that not every cell is able to proliferate to form a colony in vitro or to give rise to a tumor when transplanted in vivo. It was demonstrated afterwards that only a small fraction of the tumor cells isolated from Acute Myeloid Leukemia patients, with a characteristic marker signature, were able to establish leukemia in recipient mice. This provided a reproducible way of enriching cells with tumor-initiating activity and ruled out a Stochastic Model, which predicted that such an activity would be present in all cell fractions. Although the idea of CSCs or tumor-initiating cells had already been envisaged by

the 1960s, it was not until the identification and prospective purification of the CSCs by John Dick and colleagues, that concrete proof was provided for a Hierarchical (or stem cell) Model of cancer (Bonnet and Dick 1997), redefining cancer biology and treatment. Mouse teratocarcinoma also appeared to provide evidence for the existence of CSCs, and gave rise to a fascinating framework for studying how the cellular microenvironment contributes to oncogenesis (Damjanov 1993). The hypothesis of the CSCs theory is that cancer arises from germ cells that do not undergo normal differentiation due to specific epigenetic marks such as methylation, which normally brings about genomic instability, hypoacetylation of histones and hypomethylation of specific genes involved in the regulation of promoter regions and tumor silencing bound to oncosuppressor genes (Feinberg and Tycko 2004).

By adding halted differentiation into the definition of oncogenesis, researchers took a major step toward establishing tumor cell hierarchy as a fundamental concept in cancer biology. The signal transduction pathways associated with both normal and cancer stem cells have been under intense study (Lobo, Shimono, et al. 2007). This and other experimental results support the hypothesis that cellular heterogeneity within a tumor is part of a larger vision of cancer. Cancer is not just a simple clonal expansion of a transformed cell. It behaves like a tissue in which cells become functionally heterogeneous as a result of an aberrant differentiation of the organ itself. In this regard, tumors "act as a caricature of their corresponding normal tissues and are sustained in their growth by a pathological counterpart of normal adult stem cells" (Dalerba, Cho, et al. 2007). This might be consistent with the concept that CSCs, as for normal SCs give rise to a hierarchical organization of cell populations that underlie organogenesis (Reya, Morrison, et al. 2001). Around thirty to forty years ago, Potter (1978) somehow anticipated the idea of the involvement of epigenetic and progenitor cells/stem cells in neoplastic origin and progression. Starting from the biochemistry of cancer (1964), he envisioned neoplastic growth as a problem in intercellular communication and differentiation, and championed the concept that "oncology is blocked ontogeny" (Potter 1968, p. 587; Potter 1969). The basic idea was that cancer cells have lost a feedback control mechanism in proliferation so that their ability to divide becomes unrestricted. In addition, cancer cells acquire a variety of new properties that render them destructive to the organism as a whole. The idea that cancer might be a problem of cellular communication has also been stated by Biava (2002). Studying the relationship between cancerogens, mutagens and teratogens, his attention focused on data that showed how cancer-causing agents, when administered during pregnancy, had different effects depending on the period in which they were given. In fact, the effects manifested as an increase in malformations

if administered during organogenesis, and as an increase in the number of tumors in progeny during the period in which the formation of organs was already complete (Einhorn 1982; Lakshmi and Sherbert 1974; Brent 1980; Rice 1973; Tomatis and Mohr 1973). In the period of organogenesis, therefore, there should be regulators that impede the indefinite division of cells that is typical of malignant behavior (Biava 2008). The results obtained up to now lead to the supposition that, at least in theory, the microenvironment plays a fundamental role in the division and differentiation of both normal and tumoral cells.

This change in the levels of analysis led different authors to think that the focus of cancer research, in contrast to dominant reductionism, might consider the inherent complexity of the biological phenomena and the importance of the environment as well. Currently, there are a number of new studies indicating that the developmental limitations of tissue-specific stem cells are regulated by the microenvironment and that host cells, under specific conditions such as tissue injury or infection, might provide specific signals that counteract these restrictions (Mueller and Fusenig 2004; Nelson, DeWeese, et al. 2002). The first hint that the microenvironment is important and selective for cancer and its progression is related to the "soil hypothesis", proposed at the end of the nineteenth century by Paget (1889), Hart and Fidler (1980). The evidence that some organs provide a more fertile environment than others for the growth of certain metastases led to the belief that cancer cells show a distinct preference for different tissues. At the same time, a cancer cell merely landing in another tissue is not sufficient for a secondary tumor to develop, so that some additional properties of the tissue itself or of the tumor cell must sustain the new growth.

Different studies focused on the fate and function of stem cells, which are governed by a combination of intrinsic determinants and signals from the local microenvironment or niche, i.e., the germinal compartment in different tissues that is able to assure cellular turnover. The Epithelial Mesenchymal Transition, a program of differentiation and organization of cells mainly characterized by loss of cell adhesion, and increased cell mobility, has also been included among the mechanisms which could account for the tumor cell invasiveness (Kalluri and Weinberg 2009).

Context dependence of tumorigenicity has also been demonstrated studying the dynamic and reciprocal integration of tissue architecture and function that directs mammary gland development, tissue polarity, that ultimately drive tissue-specific gene expression. Cancer occurs when these dynamic interactions go awry for an extended time (Xu, et al. 2009). This Dynamic Reciprocity Model seems to be particularly effective in gathering molecular elements and biological processes into a powerful explanatory framework of the neoplastic process (Bissell, Radisky, et al. 2002). The

tissue organization involvement in cancer origin and progression has been eventually stated by the Tissue Organization Field Theory (TOFT). It considers cancer as a phenomemon involving tissue and not cells: carcinogenesis disrupts the tridimensional and organizational structure between the stroma and the parenchyma, mediated by cells-cells interactions, so that carcinogens might not be directly responsible for neoplasia (Sonnenschein and Soto 1999). A chain of miscommunication is a slow and subtle positive feedback of change that generates even more change. Hence, carcinogenesis and neoplasia may occur once the signals that maintain normal organization are disrupted. Thus, an understanding of the morphogenetic field paradigm (Maffini, Calabro, et al. 2005) is required to consider carcinogenesis as a developmental process gone awry (Soto and Sonnenschein 2004). During development, the temporal spatial expression of genes governs the developmental fate of cells: cells know where they came from (historical information) and where they are (positional information), and this information limits their fate to a restricted phenotype. The organism is envisioned as a society of cells and cancer represents an organizational problem within a tissue. Therefore, the need to explain the complexity of cancer through a systemic view of biological phenomena is confirmed (Bizzarri, Cucina, et al. 2008). Some philosophical considerations will follow, which arise from the above mentioned history of cancer research.

THE TWO SIDES OF THE HOURGLASS: EFFICACY OF SCIENCE AND RATIONALITY IN CANCER RESEARCH

Science is a type of knowledge that rises above simple facts, what is immediately apparent from the senses. To obtain scientific knowledge of phenomena, rationalization and experimentation are needed to validate that which is not immediately evident from ordinary knowledge. The question about causes and mechanisms involved in biological processes has been revealing a complexity that require different levels of explanation and an integrative perspective of the phenomenon itself in order to account for those functional properties which do not seem to find a satisfying explanation at the molecular level. The last mentioned theory helps us to go further in our analysis. In fact, TOFT arises from an explicit epistemological position and in opposition to the traditional SMT (Soto and Sonnenschein 2005). These researchers challenge the notion that cancer is driven by a single somatic cell that has accumulated multiple DNA mutations; stating that carcinogenesis should be considered a problem akin to normal histogenesis and tissue repair, involving the three-dimensional organization of tissues (Maffini, Soto, et al. 2004). On one hand, they review the biological presuppositions of the traditional interpretative models of cancer that considers quiescence the default state of cell prolif-

eration in metazoans, and cancer a disease of cell proliferation caused by mutations in genes controlling proliferation and the cell cycle (Sonnenschein and Soto 1999). On the other hand, they state that an organicistic, systemic, perspective has to be considered to account for the emergent properties of the higher levels of biological organization, which seem to be able to explain the neoplastic phenomena and its development , where the genome is not the driver of these processes. This new paradigm clearly contests the directives of philosophical reductionism (Soto and Sonnenschein 2004) that searches for explanation only at the lower levels of biological structures, eventually reducing the living being to molecules. In contrast, TOFT sustains that organisms are not to be explained through their elementary parts and their properties, but that a top-down causality is a more appropriate presupposition in explaining causal mechanisms when complex phenomena such as cancer are studied. In this regard, the architecture of normal tissues acquires significant importance, while mutations constitute an epiphenomenon that has no relationship to the causes of cancer. As illustrated in the following figure, the other side of the hourglass, representing the evolution of interpretative models of cancer, is in place.

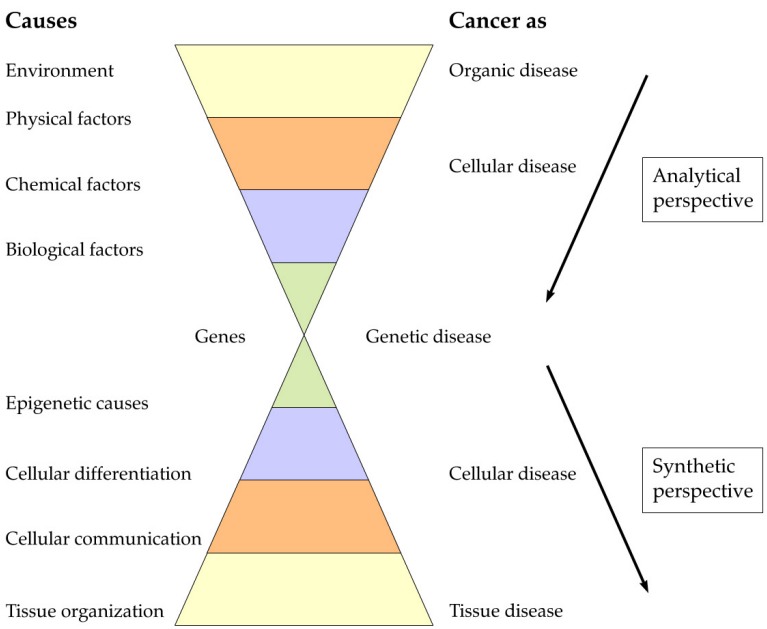

FIGURE. The two sides of the hourglass.

The efforts to explain the neoplastic phenomena had led, in the last century, to look for causes at the molecular level. Under the influence of reductionism, an analytical perspective becomes the dominant scientific approach. We have been witnesses to a process that has decomposed the phenomenon of cancer into its simple elements, identified as principle or causes that can be captured and understood immediately once isolated. Molecular biology, in this sense, has offered a suitable experimental platform for this kind of approach. Nevertheless, empirical evidence has forced the reappraisal of a more comprehensive view of the neoplastic phenomena (Bertolaso 2009), which implies a move in another direction, one that is synthetic. Once the constituent principles of living organism are identified, it takes them by default and uses them to reconstruct biological phenomena in their complexity, through explanatory argumentation (Marcos 2000).

Living organisms maintain a large variety of integral processes in a functional organization in intrinsic unity. The detailed study of these processes, typical of empirical research, necessarily reflects an analytical perspective that uncovers the natural dynamics of the process. The consolidation of experimental science during the seventieth century is due, in large part, to such application. From this standpoint, a representation of nature was derived that can be described through a simple aggregation of its components. The success of Newtonian mechanics contributed, through the triumph of mechanistic theories outlined by Descartes in the seventieth century, to this concept of nature, considered as a machine whose functioning can be explained in terms of the local movement of its parts. Stanley Jaki, in a study in which an organic vision is analyzed, as in early physics and classic mechanics (1966), noticed the genuine changes in mentality that were decisive for the development of modern science. In the hands of Galilei, Newton, and Descartes, mechanistic studies constitute a natural philosophical doctrine that has important repercussions for successive scientific developments. This did not involve a materialistic worldview—mechanistic materialism dates to the eighteenth century—but is related to a perspective in which a qualitative vision of nature is substituted with a quantitative one that can be attributed to an explicative central function from mechanical causes in the absence of final causes (Dugas 1954). An analytical point of view in science is thus an adequate means to study natural processes, implying objectivity and determining laws through mathematical tools and experimental proof. From the early stages of empirical science until the beginning of the twentieth century, such analytical method was almost exclusively used. This approach deliver enormous results starting from the structure of reality that begins with the simplest and moves toward more complex instances, and continues to provide information about many aspects that have permanent value

as they refer to a typical prospective of experimental science, which isolates different aspects of phenomena through controlled experiments.

However, such strategies have limits, as many natural phenomena cannot be explained by simple analysis of their individual components. There are in fact functional factors which manifest themselves as emergent properties that require a description and explanation from a synthetic perspective. It is the dynamism of nature and its representation that require both analytic and synthetic perspectives (Artigas 1992). A combination of this two approaches is traditionally addressed as 'systemic view' (Bunge 2004), a way of thinking about scientific problems, in our case, taking all levels of biological organization into consideration. The analytic way allows us to expand our knowledge, while the synthetic one allows us to explain phenomena already known. Even so, some philosophical foundations of the science of systems biology appear to be lacking, calling for a deeper understanding of how causality operates at different levels of organization (O'Malley and Dupré 2005).

THE CHALLENGE TO CAUSAL EXPLANATION POSED BY A MULTIPLE LEVEL PHENOMENA

The above-mentioned synthetic perspective is correlated with natural processes that implicate the appearance of new functional features and the reorganization of parts through new interactions. As the biology of cancer shows, different levels of biological complexity interacts and operates in a synergic level demanding a more comprehensive, conceptually coherent explanatory account of the neoplastic phenomena. How to integrate the different levels of causal explanation becomes a philosophical challenge which might be able to critically appraise the roles of both the analytic and synthetic accounts of causation in cancer research. In the philosophy of science literature this question has been broadly addressed in the last decades, despite a generalized disagreement among philosophers concerning the content of causal explanations in science. Thus, we will try here just to sketch out some aspects that can contribute to face this challenge. A further discussion on the different philosophical contributions is beyond the aim of this paper, although we do believe that cancer research evolution is giving us important insights on the development of the philosophy of causation in biological sciences.

Historically, the evidence of a hierarchical organization in the living systems, and in the context dependency of functional properties of its parts, has been often view as an epistemological problem: "some of these problems stem from the simple point that, in hierarchies, objects and events at the lower levels of organization *comprise* the objects and events at higher levels; but at the same time we employ different and in some

degree independent languages in the description of the different levels" (Morton 1974). Nevertheless, there is a more important philosophical challenge beyond, as stated by Woodward (2010), in elucidating the presuppositions that scientists make when distinguishing among causal relationships at different levels of biological complexity.

Usually the investigator's purpose, i.e., what is it that the researcher wishes to understand, influences the choice of level at which he works and presents explanatory evidence. Cancer research follows the same path: within the SMT the first move towards a systemic perspective, from genes to cells, is strictly related to the evidence that new elements and factors, besides genes, were causally involved in the etiopatogenesis of cancer and the need to include them in new interpretative models. TOFT is also supported by experimental evidence about the role of stroma in the maintenance of the normal functionality of epithelium (Maffini, et al. 2004). In this sense, the choice of the explanatory level is influenced by empirical consideration, so that TOFT can discard the explanatory causal role that the SMT attribute to molecular parts and address the phenomenon at the level of the tissue organization. Accordingly, it has been said "depending on the details of the case, causal description or explanation can be either inappropriately broad or general, including irrelevant details, or overly narrow, failing to include relevant detail. Which level is most appropriate will be in large part an empirical matter" (Woodward 2010).

Nevertheless, TOFT repeatedly stresses that the divergence with the SMT in explaining the neoplastic process is mainly due to their different epistemological presupposition. From the TOFT perspective, both cancer's cause and explanation reside at the tissue level of biological organization (Sonnenschein and Soto 2008). There is something new in this position. Their synthetic approach, in fact, seems to force us to reconsider the importance of the perspective adopted in explanatory argumentation when dealing with complex biological systems.

> In a general and perhaps trivial sense, molecules mediate these high-level phenomena. However, there are many interactions that occur simultaneously to maintain the structure of a tissue; hence, it is practically impossible to sort out causes and effects in a way that would precisely reveal whether emergent properties have true causal agency. Hence, biologists who take for granted that emergent phenomena exist adopt an organicist stance (Sonnenschein and Soto, 2006).

The systemic view which arises as an alternative to reductionism beyond the most general analytical approach, suggests that the multiple level explanations presented by the SMT actually require a different philosophical outlook to make sense of the unity of the phenomenon, despite the

different level it can be studied, aiming towards a comprehensive causal account of the neoplastic process as a whole. Considering carcinogenesis as an emergent phenomena which take place at higher levels of biological organization (Sonnenschein and Soto 2000), the study of cancer cannot be reduced to a complex pattern of interactions among proteins, but requires a causal explanatory definition at the level at which the pathology is observed, the tissural one, while admitting descriptive explanations at different levels.

There is an ontological presupposition beyond this position. Complex wholes are inherently greater than the sum of their parts as the properties of each part are dependent upon the context of the part within the whole in which they operate. This philosophical stance is known as organicism, or materialistic holism (Gilbert and Sarkar 2000). Von Bertalanffy (1933; 1952) viewed organicism within biology as having three major components: an appreciation of wholeness through regulation; the notion that each whole is a dynamic assemblage of interacting parts, and the idea that there are laws appropriate for each level of organization, from atoms to ecosystems. Bertalanffy's third component of organicism follows from emergent properties, which implies that different kinds of laws are appropriate for each level. Therefore, when considering an entity as complex as the cell, the fact that quarks have certain characteristics is of little relevance.

This is not to say that each level is independent of the one below; on the contrary, regularities and functional patterns [1] at one level may be dependent on those of lower levels, but they may also be dependent on the levels above it. Such is the case when considering morphogenesis, differentiation or pathologies such as cancer. These processes demonstrate that there are relational factors in nature that hold a central role within a systemic perspective and directionality. That is why the "typical theory in the biomedical sciences is a structure of overlapping interleveled causal temporal prototypical models" (Shaffner 2007) and why the analytical approach continues to provide important information about the biological systems' organization and functioning. This is also the reason why systemic accounts, which integrate an analytical approach with a synthetic one, focus their attention on the biological level at which the phenomenon acquire a specificity, taking into account its dynamic properties. In particular, meaningfulness appears, in this regard, in the already mentioned Dynamic Reciprocating Model (Ku, et al. 2009) that is presented as "the minimum required unit for expression of tissue-specific functions" which aims to expand the already existing models about the interaction between the membrane and cytoskeleton (Bissell, et al. 1982) in explaining the biological presupposition of the cancer phenotype. From a biological and explanatory point of view, it depends upon the consideration that the maintenance of a biological property is due to the activation of tissue-spe-

cific response elements. This specificity of the functional level and properties of the molecular parts, give us an important interpretative key towards an integration of the analytical and synthetic approach in a systemic outlook of biological phenomena. What has to be explained is how this specificity is maintained or missed and which causal relationships are to be taken into account to causally explain it. A "manipulationist" conception of causal explanation (Woodward 2003), from this epistemological point of view, has the advantage of fitting a wide range of scientific perspectives within the current empirical research in oncology [2]. To search for a universal level of causal description that might be the most appropriate, either at a level of maximal detail or, on the contrary, at a privileged level of maximal generality or abstractness, seems not to be adequate in biology. The challenge to find the right level of causal explanations of biological phenomena seems to demand for a deeper understanding of the mechanisms and principles which characterize the complexity of living systems and their biological meaning.

That is why "a more holistic, hierarchical approach to carcinogenesis (...) yields many observations that are difficult to explain from a purely reductionist perspective" (Root-Bernstein, 1999). Interpretative models which build the first side of the hourglass (cf. figure) have not led to a unitary vision of cancer, because an analytical reductionist approach does not allow us to identify the right level of regularities in the neoplastic phenomenon that might consent a unique definition. What emerges, in contrast, is the evidence of causal complexity, which implies organizational and structural dynamics. It requires the addition of a different ontological outlook, like organicism, and a synthetic epistemological perspective, to make sense of all the empirical evidence already in our hands. Once the level of inquiry is identified, systemic perspectives are ready to move from a level of organization to a lower one in order to explore different, molecularly described aspects of the biological organization, avoiding the risk of a causal reductionist explanation of the phenomenon.

Dynamics and interactions have been also described within the reductionist paradigm, but this enlargement in the perspective is just basically attributed to a greater number of molecular factors that interact with each other, so that intrinsic limits of reductionism cannot be overcome. The genetic explanation of cancer still remain paradigmatic within those models, where significant paradoxes arise, that reductionism does not seem to deal with (Baker and Kramer 2007). Nonetheless, many molecular biologists, and some philosophers, would hold that the road to wider explanatory power, greater predictive precision, and an ever-increasing payoff in reliable technological applications, is paved by reduction. This is taken to the extent that philosophers who have concerned themselves with ap-

plied science often have seen it as primarily focused on prediction, and failed to appreciate how different prediction is from control.

It can be argued, that if there are obstacles in the reduction of the biological to the macromolecular, they are temporary or at least do not represent either logical or physical obstacles; reduction is still for the most part, the most powerful means to correct, deepen, and broaden scientific theory (Rosenberg 2006). Although reductionism still has unquestionable utility—especially when, due to of its analytical approach, it has represented an effective way to make technology reliable enough to employ—in our opinion complexity in biology is something that cannot be understood by simplifying things, by just reducing their causal explanation through dissection and analysis. Cancer seems to reveal this biological complexity, and the recent history of cancer research could be a precious case study for philosophical reflection. It has been observed that in their enthusiasm for reductive accounts,

> philosophers have often misdescribed or oversimplified the content of the causal and explanatory claims they have hoped to reduce. We need more careful description of just what such claims say (and of the regularities and counterfactuals associated with them). Only after this has been done should we investigate what sorts of reductionisms are possible (Woodward 2003, p. 22).

Functions of individual parts, their biological meaning and evidence of dynamics in the neoplastic process, transcend the properties of the molecules involved, shaking the foundations of the dominant reductionist position. A synthetic approach and a systemic outlook of cancer are required to couple with the double nature of biological phenomena: its material component and its integrated functional properties.

CONCLUSIONS

From an epistemological and philosophical point of view we are at a very interesting position, if we accept, as in daily laboratory work, that the big picture can never be missed. Somehow, the analytical approach, beyond the dominant reductionism, has provided the necessary elements for the elaboration of successive theories, and might be compared to the grains of sand in an hourglass, while organicism can be considered like the hourglass itself, powered by the sand within. Reductionism has created an oscillation in the attribution of a causal role among different kinds of molecules and the processes involved, while organicism has provided explanations of cancer identifying a specific level at which the properties of tumors have to be addressed and understood through a synthetic approach. Although we think that organicism is an advance, the ultimate test will come from usefulness: whichever of the two perspectives can best

integrate the notions derived from the other will be more able to interpret biological phenomena in all its complexity. In fact, there is no experimental design that is completely independent from theoretical interpretation, as application and control of a scientific theory is not automatic and requires a certain amount of creativity and interpretation.

Experimental research depends on the capacity to comprehend, which goes beyond simple observation and compilation of data. Scientists normally take for granted that we live in a rational and ordered cosmos which is subject to precise laws that can be discovered through reasoning. Even if the scientific method works, the success of discoveries about nature's dynamics continues to be the object of intense discussion (Davies 1993, p. 20). Our case study suggests that different perspectives and their integration might allow us to better understand how functional order/disorder of living beings shall be addressed. The complexities of biological causality do not justify embracing non-scientific ideologies, such as vitalism, but "should encourage all those who are dealing with them to give a broader basis to the concept of causality" (Mayr 1988).

The relationship between theory and experience is crucial in defining a conceptual framework that will permit a more adequate understanding of cancer. If "the competition between paradigms is not the sort of battle that can be resolved by proofs" (Kuhn, 1962), we still need to consider that science is reason in action. That is why it has been stated that if the power of human rationality is threatened, the value of science is at risk, and if rationality itself is threatened, then the situation becomes absurd (Trigg 1993, pp. 220-221). On the other hand, a deeper knowledge of the mechanisms that have driven experimental research over the last century shall allow us to avoid this risk. Limits of reductionism and questions posed by organicism can thus offer new opportunities for theoretical science in biomedicine (Yun 2008). It is our belief that the time is ripe to establish this challenge, but only the next years will reveal if the challenge will be accepted.

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NOTES

1 We rather avoid using the term *law* here, taking into account the huge debate in the philosophy of biology about the real existence of laws in biology, while it seems to be more appropriate to use the concept of regularities to address some explanatory features and object of inquiry (cf. cf. Ayala F. J., Dobzhansky (Ed). *Studies in the Philosophy of Biology*. University of California Press, Great Britain 1974; Rosenberg A, McShea D. *Philosphy of Biology*. Routledge, New York 2008; Fox Keller E. "It is possible to reduce biological explanations to explanations in chemistry and/or physics?" In *Contemporary Debates in Philosophy of Biology*. Edited by J. Ayala and R. Arp. Wiley-Blackwell, 2010, beside Woodward and Shaffner's cited in References.).

2 Woodward has emphasized in various ways how important our ability to intervene and manipulate nature is in the development of a scientific understanding of nature. However a further discussion about how this theory of causal explanation can account for the different methodological approaches in cancer research is missed here, requiring a wider presentation of empirical data from scientific literature.

REFERENCES

Artigas, M. (1992), *La inteligibilidad de la natural*eza, EUNSA, Pamplona, p. 87.

Baker, S. G., Kramer B. S. (2007), "Paradoxes in carcinogenesis: new opportunities for research directions", *BMC Cancer* 7: 151–157.

Bertolaso M. (2009), "Towards an integrated view of the neoplastic phenomena in cancer research," *Hist. Phil. Life Sci*. 31: 79-98.

Biava, P.M. (1999), "Complexity and cancer," http://www.cesil.com/cesil99/biavaeng.htm (Accessed March 2008).

Biava, P. M. (2002,) *Complessità e biologia. Il cancro come patologia della comunicazione*, Milano: Monadori.

Bissell, M. J., Hall, H. G., Parry, G. (1982), "How does the extracellular matrix direct gene expression?" *J. Theor. Biol*. 99: 31-68.

Bissell, M. J., Radisky, D. C., Rizki, A., Weaver, V. M., Petersen, O. W. (2002), "The organizing principle: microenvironmental influences in the normal and malignant breast," *Differentiation* 70: 537-46.

Bizzarri, M., Cucina, A., Conti, F., D'Anselmi, F. (2008), "Beyond the Oncogene Paradigm: Understanding Complexity in Cancerogenesis," *Acta Bioth*. 56: 173-196.

Bonnet, D., Dick, J. E. (1997), "Human acute myeloid leukaemia is organized as a hierarchy that originates from a primitive hematopoietic cell," *Nature Med*. 3: 730-737.

Boveri, T. (1914), "Concerning the origin of malignant tumours," *Journal of Cell Science* (2008) 121: 1-84.

Brent, R. L. (1980), "Radiation teratogenesis," *Teratology,* 21: 281-298.

Bunge, M. (2004), *Emergence and Convergence*, Toronto: University of Toronto Press, pp. 40-52.

Burmeister, T. (2001), "Oncogenic retroviruses in animals and humans," *Rev. Med. Virol*. 11: 369-80.

Dugas, R. (1954), *La mécanique au XVIIe siècle*, Paris: Neuchatel, Le Griffon.

Colditz, G. A., Sellers, T. A., Trapido, E. (2006), "Epidemiology identifying the causes and preventability of cancer?," *Nat. Rev. Cancer* 6: 75.

Comings, D. E. (1973), "A general theory of carcinogenesis," *Proc. Natl. Acad. Sci.* USA 70:3324-3328.

Cooper, G. M., Okenquist, S., SiLverman, L. (1980), "Transforming activity of DNA of chemically transformed and normal cells," *Nature* 284: 415-421.

Dalerba, P., Cho, R. W., Clarke, M. F. (2007), "Cancer stem cells: models and concepts," *Ann. Rev. Med.* 58: 267-84.

Damjanov, I. (1993), "Teratocarcinoma: neoplastic lessons about normal embryogenesis," *Int. J. Dev. Biol.*37: 39-46.

Davies P. (1993), *The Mind of God. The Scientific Basis for a Rational World*, New York-London: Simon and Shuster.

Doolittle, R. F., Hunkapiller, M. W., Hood, L. E., Devare, S. G., Robbins, K. C., Aaronson, S. A., Antoniades, H. N. (1983), "Simian sarcoma virus one gene, v-sis, is derived from the gene (or genes) encoding a platelet-derived growth factor," *Science* 221: 275-277.

Einhorn, L. (1983), "Are there factors preventing cancer development during embryonic life?," *Oncodev. Biol. Med*. 4: 219-229.

Fearon, E. R., Vogelstein, B. (1990), "Genetic model for coloreotal tumorigenesis," *Cell* 61: 759-767.

Feinberg, A. P., Ohlsson, R., Henikoff, S. (2006), "The epigenetic progenitor origin of human cancer," *Nature Reviews Genetics* 7: 21-33.
Feinberg, A. P., Tycko, B. (2004), "The history of cancer epigenetics," *Nature Rev. Cancer* 4: 143–153.
Feinberg, A. P., Vogelstein, B. (1983), "Hypomethylation distinguishes genes of some human cancers from their normal counterparts," *Nature* 301: 89-92.
Furth, J., Kahn, M. C. (1937),"The transmission of leukaemia of mice with a single cell," *Am. J. Cancer* 31: 276-282.
Gatenby R. A., Vincent, T. L. (2003), "An evolutionary model of carcinogenesis," *Cancer Res.* 63: 6212-20.
Gatenby, R. A., Frieden, B. R. (2002), "Application of information theory and extreme physical information to carcinogenesis," *Cancer Res*. 62: 3675-84.
Gilbert S. F., Sarkar, S. (2000), "Embracing complexity: organicism for the 21st century," *Developmental Dynamics* 219: 1-9.
Harris, H., Rawlins, J., Sharps, J. (1996), "A different approach to tumour suppression," *J. Cell. Sci.* 109: 2189-2197.
Harris, M. (1971), "Cell fusion and the analysis of malignancy," *Proc. R. Soc. Lond. Biol. Sci.* 179: 1-20.
Hart, I. R., Fidler, I. J. (1980), "Role of organ selectivity in the determination of metastatic patterns of B16 melanoma," *Cancer Res.* 40: 2281-2287.
Jaki, S. L. (1966), *The Relevance of Physics*, Chicago: Chicago University.
Jones, P. A., Baylin, S. B. (2007), "The epigenomics of cancer," *Cell* 128: 683.
Jones, P. A., Taylor, S. M. (1980), "Cellular differentiation, cytidine analogs and DNA methylation," *Cell* 20: 85-93.
Kalluri R, Weinberg RA. (2009), "The basics of epithelial-mesenchimal transition," *J. Clin. Invest.* 119: 1420-8.
Kitano, H. (2005), "Cancer as a robust system: implications for anticancer therapy," *Nature reviews/System. Biology,* May 2005, p. 47.
Klein, G. (2002), "Perspectives in studies of human tumor viruses," *Front. Biosci.* 7: d268.
Knudson, A. G. Jr. (1971), "Mutation and cancer: statistical study of retinoblastoma," *Proc. Nat. Acad. Sci. USA* 68: 820-823.
Kuhn, T. S. (1962), *The Structure of Scientific Revolutions*, Chicago: Chicago University.
Lakshmi M. S., Sherbert, G. V. (1974), *Embryonic and Tumor Cells Interactions*, NY: Karger, pp. 380-399.
Land, H., Parada, I. F., Weinberg, R. A. (1983), "Tumorigenic conversion of primary embryo fibrobiasts requires at least two cooperating oncogenes," *Nature* 304: 596-602.
Lloyd, A. C, Obermüller, F., Staddon, S., Barth, C. F., McMahon, M., Land, H. (1997), "Cooperating oncogenes converge to regulate cydin/cdk complexes," *Genes Day* 11: 663-677.
Lobo, N. A., Shimono, Y., Qian, D., Clarke, M. F. (2007), "The biology of cancer stem cells," *Annu. Rev. Cell. Dev. Biol*. 23: 675-99, Table 1.
Luria, S. (1960), "Viruses, cancer cells, and the genetic concept of virus infection," *Cancer Res*. 20: 677-88.
Maffini, M. V., Calabro, J. M , Soto, A. M., Sonnenchein, C. (2005), "Stromal regulation of neoplastic development: age-dependent normalization of neoplastic mammary cells by mammary stroma," *Am. J. Pathol*. 167: 1405-1410.

Maffini, M. V., Soto, A. M., Calabro, J. M., Ucci, A. A., Sonnenschein, C. (2004), "The stroma as a crucial target in rat mammary gland carcinogenesis," *J. Cell. Sci*. 117: 1495–1502

Marcos A. (2000), *Hacia una filosofia de la ciencia amplia,* Madrid: Tecnos.

Mayr, E. (1988), *Towards a New Philosophy of Biology,* Cambridge: Cambridge University Press.

Morton, B. (1974), "Reduction, hierarchies and organicism," in *Studies in the Philosophy of Biology*, Ayala, F. J. and Dobzhansky, T. (Eds.), LA: University of California Press.

Mueller, M. M., Fusenig, N. E. (2004), "Friends or foes—bipolar effects of tumour stroma in cancer," *Nature Rev. Cancer* 4: 839–849.

National Toxicology Program (2005), *Report on Carcinogens*, 11th ed., Bethesda Similarities, MD: US Dept of Health and Human Services, PHSNTP.

Nelson, W. G., De Weese, T. L., De Marso, A. M. (2002), "The diet, prostate inflammation, and the development of prostate cancer," *Cancer Metastasis Rev*. 21: 3–16.

Nowell, P. C. and Hungerford, D. A. (1960), "A minute chromosome in human chronic granulocytic leukaemia," *Science* 132: 1488-1501.

O'Malley, M. A. and Dupré, J. (2005), "Fundamental issues in systems biology," *Bioessays* 27: 1270-1276.

Paget, S. (1889), "The distribution of secondary growths in cancer of the breast," *Lancet* 1: 571-573.

Parkin, D. M. (2004), "International variation," *Oncogene* 23: 6329.

Potter, V. R. (1964), "Biochemical perspective in cancer research," *Cancer Research* 24: 1085-98.

Potter, V. R. (1968), "Mechanisms of carcinogenesis in relation to studies on minimal deviation hepatomas," in *Exploitable Molecular Mechanisms and Neoplasia,* Austin, p. 587.

Potter, V. R. (1969), "Recent trends in cancer biochemistry: the importance of studies on fetal tissues," *Proc. Can. Cancer Conf*. 8: 9-30.

Potter, V. R. (1978), "Phenotypic diversity in experimental hepatomas: the concept of partially blocked ontogeny," The 10th Walter Hubert Lecture, *Br. J. Cancer* 38: 1-23.

Reya, T., Morrison, S. J., Clarke, M. F., et al. (2001), "Stem cells, cancer, and cancer stem cells," *Nature* 414: 105-111.

Rice, J.M. (1973), "An overview of transplacental chemical carcinogenesis," *Teratology* 8: 113-125.

Rosenberg, A. (2006), *Darwinian Reductionism*, Chicago: Chicago University Press.

Root-Bernstein, R. S. (1999), "Complementarity and contradiction in cancer research: The role of hierarchies in carcinogenesis," *Anticancer Research* 19: 4915-18.

Rous, P. (1910), "A transmissible avian neoplasm (sarcoma of the common fowl)," *J. Exp. Med*. 12: 696-705.

Ruley, H. E. (1983), "Adenovirus early region 1A enables viral and cellular transforming genes to transform primary cells in culture," *Nature* 304: 602-606.

Schaffner, K. F. (2007) "Theories, models and equations in systems biology," in *Systems Biology. Philosophical Foundations,* Boogerd F. C., Bruggeman F. J., Hofmeyer J. H. S., Westerhoff H. V. (Eds.), NY: Elsevier.

Shih, C., Shilo, B. Z., Goidfarb, M. P., Dannenberg, A., Weinberg, R. A. (1979), "Passage of phenotypes of chemically transformed cells via transfection of DNA and chromatin," *Proc. Natl. Acad. Sci. USA* 76: 5714-5718.

Shih, C., Weinberg, R. A. (1982), "Isolation of a transforming sequence from a human bladder carcinoma cell line," *Cell* 29: 161-169.

Sonnenschein, C., Soto, A. M. (1999), *The Society of Cells: Cancer and Control of Cell Proliferation*, NY: Bios Scientific.

Sonnenschein, C., Soto, A. M. (2000), "Somatic mutation theory of carcinogenesis: Why it should be dropped and replaced," *Molecular Carcinogenesis* 29: 205-211.

Sonnenschein, C. and Soto A. M. (2006), "Carcinogenesis and metastasis now in the third dimension—what's in it for pathologists?," *American Journal of Pathology* 168: 363-366.

Sonnenschein, C. and Soto, A. M. (2008), "Theories of carcinogenesis: An emerging perspective," *Seminars in Cancer Biology* 18: 372-377.

Soto, A. M., Sonnenschein, C. (2004), "The somatic mutation theory of cancer: Growing problems with the paradigm?," *BioEssays* 26: 1097-1107.

Soto, A. M., Sonnenschein, C. (2005), "Emergentism as a default: Cancer as a problem of tissue organization," *J. Biosci*. 30: 103-18.

Steel, D. M., Harris, H. (1989), "The effect of antisense RNA to fibronectin on the malignancy of hybrids between melanoma cells and normal fibroblasts," *J. Cell. Sci.* 93: 515–524.

Tomatis, L., Mohr, V. (Eds.) (1973), *Transplacental Carcinogenesis*, Lyon: IARC Sci. Publ.

Trigg, R. (1993), *Rationality and Science. Can Science Explain Everything?,* Oxford: Oxford Univerity Press.

Vogelstein, B., Kinzler K.W. (2004), "Cancer genes and the pathways they control," *Nature Medicine* 10: 789-799.

Von Bertalanffy, L. (1933), *Modern Theories of Development* (J. H. Woodger, trans.), NY: Harper.

Von Bertalanffy, L. (1952), *Problems of Life*, NY: Harper.

Waterfield, M. D., et al. (1983), "Platelet-derived growth factor is structurally related to the putative transforming protein p28sis of simian sarcoma virus," *Nature* 304: 35-39.

Woodward, J. (2003), *Making Things Happen—A Theory of Causal Explanation*, Oxford: Oxford University Press.

Woodward, J. (2010), "Causation in biology: stability, specificity, and the choice of levels of explanation," *Biology and Philosophy* 25 (3): 287-318.

Xu R., Boudreau A., Bissell M. J. (2009), "Tissue architecture and function: dynamic reciprocity via extra- and intra-cellular matrices," *Cancer Metastasis Rev*. 28: 167-76.

Yun, A. J. (2008), "The hegemony of empiricism: the opportunity for theoretical science in medicine," *Med. Hypotheses* 70: 478-81.