What Mechanisms Can't Do: Explanatory Frameworks and the function of the p53 in molecular oncology

1. Introduction

Thinking about mechanisms is part and parcel of the philosophy of science agenda in the last decade. What has been called "the new mechanistic philosophy" (Skipper and Millstein 2005) conceives of mechanisms as the main providers of biological explanation. The latter is, in order, an open-ended endeavour mainly consisting in filling in the missing causal links of a biological mechanism. Even if we have not much to argue against this view in itself, we think it tells only part of the story concerning explanations in the life sciences. In particular, "the new mechanistic philosophy" describes mechanisms, and their use by practising scientists, as the only necessary and sufficient providers of scientific explanations[1]. Drawing on examples in molecular oncology, we will show that explaining complex biological phenomena (cancer, in our case) requires instead a dynamic interaction between the mechanistic level – rendered at the appropriate degree of ontological resolution – and far more general explanatory tools that perform a fundamental epistemic role in the provision of biological explanations. We call such tools "explanatory frameworks". They are called *frameworks* to stress their higher level of generality with respect to bare mechanisms; on the other hand, they are called *explanatory* because, as we show in this paper, their importance in explaining biological phenomena is not secondary with respect to mechanisms.

The account we propose complements standard mechanistic approaches by making them more complete and dynamic. It will make them more complete because our approach does not take into account mechanisms as the only providers of relevant explanations. Rather, we conceive of mechanisms as relevant to explanation only insofar as they are embedded into broader explanatory frameworks telling what mechanisms are to be searched and described in order to account for a biological phenomenon. Furthermore, our account is more dynamic because it is able to better explain both continuities and discontinuities in biomedical science. We are able to explain discontinuities by pointing to different explanatory frameworks that give significance to mechanisms. We show that scientific change, in the context of molecular oncology, implies the integration of old mechanisms into, and by means of, new explanatory frameworks that characterize scientific change. Mechanistic accounts fail to do this because they mostly concentrate on the way mechanisms are constructed: they can at best conceive of scientific progress as a process of adjustment and revision of mechanistic modules.

In this paper, we start by recalling briefly the main tenets of the mechanistic epistemology

1 See Craver (2007) where mechanisms are explicitly taken to be the *explanans* of biological phenomena.

(section 2) and its treatment of the problem of relevance (section 2.1). We argue that something is missing in this account of scientific work, and propose the notion of explanatory framework to fill this gap (section 3). We then illustrate our account through a case study taken from molecular oncology, therein we show the function explanatory frameworks fulfil in mechanisms discovery and explanation (section 4). We show how explanatory frameworks establish selective and local criteria of causal relevance that drive the search for, characterization and use of biological mechanisms during the rise of the so-called "oncogene paradigm" in the late Seventies and the discovery of gene p53 (4.1 and 4.2). We then further characterize explanatory frameworks with respect to their explanatory function through the example of the emergence of the idea that stem cell biology may explain peculiar characteristics of carcinogenesis and tumor recurrence: we show that explanatory frameworks allow for changes of scientific perspective on the causal relevance of mechanisms without necessarily fully replacing previous explanatory frameworks, nor through the simple refinement or discovery of mechanisms (4.3 and 4.4). Moreover, we show that explanatory frameworks can steer scientific discovery in some directions rather than others: the further mechanistic elucidation (functional characterization) of the role of tumor suppressor gene p53, for instance, is made possible exactly by the emergence of the new explanatory framework of cancer stem cells (4.4). The new functional implication of p53 in regulating symmetric-asymmetric cell division was not visible under the previous explanatory framework (i.e. the oncogene paradigm), therefore arguing that the new explanatory framework (i.e. the cancer stem cell hypothesis) enables a novel mechanistic explanation of certain features of cancer cells. Finally, we use our case study to recapitulate the features of explanatory frameworks and their epistemic role in an attempt to offer a more complete picture of our account; we discuss broader issues connected to their relationship with other philosophical account of mechanism discovery (section 5), and we offer some concluding remarks on the issues touched in this paper.

2. Mechanicism[2]

Explanation, according to the long dominant logical empiricist account, amounts to an argument showing that the occurrence of a given phenomenon (*explanandum*) could have been expected on the basis of laws of nature, knowledge of background conditions or premises and the application of valid rules of inference (Hempel 1965). This model has been criticised in many ways

2 In the remainder of this paper we will follow a nomenclature proposed in a recent paper (Nicholson 2012, 154-5). When referring to the philosophy of science account of explanation in terms of mechanisms, we will therefore use the adjective "mechanismic" instead of the more frequent "mechanistic" (or "mechanicistic") to stress, as proposed by Nicholson, that we only refer to the epistemological part of the philosophical program originally advanced by Machamer, Darden and Craver (2000). Since in this paper we focus on the causal role of mechanisms for a philosophy of explanations, we refrain from discussing the metaphysical foundations of "mechanistic" philosophy that, as Nicholson argues, have a different and much longer philosophical history than the concept of "mechanism" we discuss here.

and does no longer enjoy much support in philosophy of science (for a review of these debates, see Salmon 1989). Among the proposed alternative accounts, several authors emphasized the explanatory role of causation (Lewis 1986, Salmon 1984, Woodward 1984). In these accounts, explaining a phenomenon amounts to giving information on the antecedent causes that produced it, like in the very trivial case of explaining the presence of smoke by the necessary presence of fire. The aetiological focus of these accounts thus downplayed the predictive role of laws and law-like conceptualizations in providing explanations. The aetiological account of explanation was recently revised by the proponents of the mechanismic view. According to Craver (2007) for example, the explanation of a phenomenon occurs when the network of entities and activities (i.e. the mechanism) that produce it are appropriately described. In his view explanations are therefore finally accounted for in terms of mechanisms: mechanisms are the *explanans* (Craver 2007, 22 & 139) of a given phenomenon and, so to say, perform the job of revealing its causal structure (*ivi*, 27).

In the last decade a great number of philosophers have started to analyse the concept of "mechanism" as a fruitful meta-scientific notion that captures what the actual activity of practising life scientists is about when they are after explaining the inner workings of biological phenomena. Within the remit of the mechanismic program, philosophers have studied both what "mechanism" and "mechanistic explanation" of a given phenomenon mean (Bechtel 2006; Bechtel and Abrahamsen 2005; Glennan 2005; Glennan 2002), and the epistemic strategies to the discovery of mechanisms in the real world (Bechtel and Richardson 1993; Craver 2007, Darden 2002, Darden and Craver 2002). According to the characterization provided by Machamer, Darden and Craver in their seminal paper in this field

> "Mechanisms are entities and activities organized such that they are productive of regular changes from start or set-up to finish or termination conditions" (Machamer et al. 2000, 3).[3]

Mechanismic philosophers also provide an epistemic analysis of mechanisms, by describing a set of reasoning strategies that aid their discovery and refinement. Darden (2002), for instance, put forth three of these strategies: schema instantiation, modular subassembly, and forward/backward chaining[4]. The mechanism of protein synthesis is one classical example used to illustrate the kind of reasoning process Darden and Craver (2002) have in mind. In their reconstruction, the general

3 Bechtel and Abrahamsen give a similar characterization: "A mechanism is a structure performing a function in virtue of its component parts, component operations, and their organization." (Bechtel & Abrahamsen 2005, 423) Of course, authors differ in the details of their account. However, since our main aim here is to argue against the explanatory monism of mechanicism, entering the minute detail of these differences is unnecessary.

4 "Schema instantiation provides an abstract type of mechanism that may be specified to apply to a particular case. Types of mechanisms may be depicted in abstract mechanism schemata; instantiation is the process of making a schema less abstract and applicable to a particular case. In modular subassembly one searches for type of modules to assemble into a hypothesized mechanism. A strategy operating at even finer grain is to reason stage by stage about how gaps in what is known about the productive continuity of a mechanism are to be filled, either forward chaining from a convenient starting point or backward chaining from a later stage" (Darden, 2002).

schema of protein synthesis (DNA→RNA→Protein) provides the abstract mechanism that then needs to be instantiated in specific circumstances. As scientific enquiry proceeds, so the reconstruction goes, scientists divide up the mechanism into more restricted modules that should explain how a particular activity of the general mechanism is performed (for instance, transcription of genetic information into mRNA). At an even finer level of resolution, scientists operate back and forth from the abstract schema to mechanisms to further specify the kind of entities and activities involved in the production of the phenomenon of interest. In this view, scientific progress occurs through a gradual refinement of the mechanism responsible for the phenomenon. Sometimes this process is presented as an endeavour that aims "[…] to transform black boxes (components and their function unknown) to gray boxes (component functions specified) to glass boxes (components supported by good evidence)" (Darden 2008, 967). Bechtel and Abrahamsen give a similar account:

> "research typically begins with an oversimplified account in which only a few components and aspects of their organization are specified. Over time, it is repeatedly revised and filled in [...] Much of the discovery and testing involved in mechanistic explanation focuses on proposing components or forms of organization that are to be added to or used to revise parts of a sketch" (Bechtel and Abrahamsen 2005, 436-437)

In this ideal frame, mechanisms are piecemeal constructed and, after some time and amassing of evidence, they are delivered to textbooks and are no longer the focus of research projects. Although we recognize that such a re-construction can account, in hindsight, for much explanatory endeavours in the life sciences, we argue that its suggested linearity only partially illuminates the strategies of mechanism discovery and their role in explanation of complex biological phenomena like, for instance, cancer. We will return to this issue in sections 3 and 5 when we will emphasize how our account of mechanism discovery is different from existing ones.

Overall, we believe that the recent interest for the epistemic role of biological mechanisms was driven by a laudable descriptive engagement with the actual activity of practising life scientists. However, this strand of philosophical analysis should avoid concentrating on mechanisms as the exclusive source of explanation and locus of scientific work. In particular, as we show in the next section, such view seems not to do justice to the actual way in which the life sciences are practised, thus undermining the original descriptive intention of the mechanismic program.

What we regard as problematic in the mechanismic account is that, it only poorly accounts for how certain entities and activities acquire (and change) explanatory relevance, and how certain mechanisms and not others become the focus of experimental activities in a given field of life sciences. In the next section, we briefly present the problem of relevance and its treatment by mechanismic philosophers. We claim that alone, the mechanismic account is insufficient to answer these questions. Coupling it with the concept of explanatory framework, we suggest, might fill this gap.

2.1. The problem of relevance

All elements in the causal history of a phenomenon are not equal. As Craver writes,

> "Providing an etiological explanation involves not merely revealing the causal nexus in the past light cone of the *explanandum* phenomenon. It involves, in addition, selecting the relevant interactions and processes and picking out the relevant features of those processes and interactions." (Craver 2007, 78)

Craver recalls a very classical example by Hitchcock (1995), initially used to discuss Salmon's account of causality: using a pool cue, one hits a pool ball and imparts it with a linear momentum. However, since there is also chalk on the tip of the cue, the act also imparts it with a blue mark. By Salmon's criteria, both elements are causal. Hitchcock's point is that while the momentum is relevant to the trajectory of the ball, the blue mark is not. Craver (2007) relies on a manipulationist account of causation (Woodward 2003) to solve this general kind of problem: while one can manipulate the trajectory by manipulating the momentum, one cannot do the same by manipulating the blue mark.

As a matter of fact, the blue mark might well be relevant to the trajectory of the ball. If we are precise enough in our measurements, even a very small amount of chalk will make a difference to the trajectory. Nevertheless, there is an important sense in which the chalk is irrelevant, and it depends on the context of explanation: because we are dealing with very small amounts of chalk, and because we are not interested in very minor changes in trajectory (pool players are generally interested in whether they sink the ball or not), the chances of the chalk *making a difference* to the phenomenon are too small to be of any practical relevance. Therefore, while the full causal history would include both the momentum and the chalk, the mechanismic explanation (rightfully) excludes the latter. This is not an ontological claim about the blue mark, but only a claim about the "normal range" in which the mass of chalk varies and the differences that are relevant to the pool player – highly pragmatic considerations. As Craver notes:

> "The causal relevance relations under different ranges of conditions are objective features of the world. However, which of those objective relations is relevant depends on what you are trying to explain." (Craver 2007, 100)

In principle, this could be done objectively: if we could measure actual variations of chalk and their actual effect on whether the ball sinks or not, we could claim objectively that the chalk has very little relevance (at least in situations similar to those studied). However, this proves harder than it seems, especially in the life sciences, where phenomena are considerably more complex than a game of pool. A high number of elements will inevitably end up having "some relevance", and having even very little relevance is not being irrelevant, especially considering that we study only a

small subset of the conditions in which a given phenomenon operates. Craver dismisses this as a practical difficulty that we should try to overcome:

> "But these practical difficulties, which are part of what make science challenging and rewarding, do not impugn the overall idea that what one ideally wants to establish is precisely such well-controlled relationships of manipulability." (Craver 2007, 103)

Although one might agree that scientists aim for this ideal, it is generally quite far from reach. Any attempt to establish what components are necessary and sufficient for a mechanistic explanation will face the intrinsic difficulty that relevance comes in degrees, and that in general these degrees are not well-known at all. Contrary to mechanismic philosophers, we do not consider this a technical problem, but rather an essential feature of scientific activity. Understanding scientific developments, we claim, implies understanding why, in a given period and with incomplete knowledge, some parts of the causal history were deemed more relevant than others. While mechanisms might be an essential part of an account of scientific change, they tell only part of the story.

In this paper, we speak of causal relevance in a phenomenal way, that is, with reference to the actual importance that a causal explanation acquires within a given set of practices in the life sciences – in our case, molecular oncology. What scientists consider relevant is, at best, an approximation of a more ontic relevance. But it would be a mistake to dismiss the discrepancy between the two as mere ignorance waiting to be dispelled. It is the very conditions in which science is practised, and therefore it is necessary for history and philosophy of science to account for the way scientist deal with it.

Bechtel and Richardson (1993) have devoted considerable attention to scientific discovery, discussing the different constraints (social, theoretical, empirical, and cognitive – with a particular emphasis on the last two) that drive the discovery of mechanisms. They are especially concerned with the identification of a *locus of control*, which according to them "evolves with time and research as scientists develop conceptual frameworks to determine a particular way of decomposing nature into systems." (Bechtel and Richardson 1993, 40) However, like in Craver's account (2007), they consider the identification of a locus of control to be clear-cut and exclusive: "the decision virtually defines a research program or tradition, and a resolution will preclude alternative lines of inquiry." (Bechtel and Richardson 2000, 40) As will become clear with the example of cancer, we wish to highlight that the different parts of the aetiology of a phenomenon become differentially relevant in a much more diffuse way: rather than "making it or not" in the mechanism, they gradually come to be seen as more or less important to its understanding. An account of either scientific change or explanation needs to make conceptual sense of these movements and of the kind of rationalization grounding them. To this aim we propose the notion of explanatory frameworks (section 3), which will be further characterize and given empirical warrant by means of

a case study taken from molecular oncology (section 4).

3. Explanatory Frameworks

The easiest way to characterize what we have in mind with the expression "explanatory framework" is through an example. One straightforward instance of explanatory framework is the so-called "oncogene paradigm". With this expression, historians of science, and scientists as well, refer to a pattern of explanation of the cause(s) of cancer having to do with the driving genetic role of specific genes (oncogenes and tumor suppressors[5]). According to the oncogene paradigm, cancer is caused by altered expression and/or genetic modification of a specific set of genes. Point mutations, retroviral insertions and major chromosomal alterations can increase the expression of specific genes (oncogenes - OG) that transform the biological activity of a cell in a dominant fashion, thus rendering it tumorigenic. On the other hand, the same kinds of alteration may lead to the repression of transcription – typically in a recessive or dominant negative fashion – thus suppressing the protective activity of other genes (tumor suppressors - TS) and eventually causing the malignant transformation of the cell. In current versions of this explanatory framework, it is assumed that cancerous transformation is a multistep process (Vogelstein and Kinzler 1993): it thus takes a series of genetic alterations in OGs and TS genes to overcome cellular mechanisms of DNA repair, start the malignant transformation and sustain the proliferation and propagation of cancer cells. Finally, OGs and TS genes exert their activity through intricate pathways of signal transduction whose alteration ultimately confers the aggressive malignant phenotype that is typical of cancer cells.

In the jargon of both historians and practising scientists, explanatory frameworks are often referred to with a number of expressions that share some "family resemblance" (Wittgenstein 1953) with our "explanatory framework". Molecular oncology offers many examples of this variety, including the use of wordings like "paradigm" (Bishop 1995; Morange 1997), "vision" (Morange 1997), "theory" (Sonnenschein and Soto 2008), "model" (Temin 1974, Vogelstein and Kinzler 1993), "metaphor" (Hanahan and Weinberg 2000), "perspective" (Breivik and Gaudernack 1999), "concept" (Maenhaut et al. 2010), "hypothesis" (Donnenberg and Donnenberg 2005), up to "theory-method package" (Fujimura 1988, Fujimura 1992). Each of these labels being loaded with philosophical meaning, we would like to make some conceptual order into this rather uncontrolled proliferation of uses. We are interested in the specific task of understanding the relations between

5 In this paper we take the oncogene paradigm to include the activity of both oncogenes and tumor suppressors as aetiological explanations of cancer. To be sure, however, oncogenes where discovered before tumor suppressors and actually drove the realization that cancer, long believed to be mainly due to dysregulation and viral infection, is instead best accounted for as a full-fledged genetic and epigenetic disease (see Keating and Cambrosio 2012; Keating, Cambrosio and Mackenzie 1992; Morange 1997).

the mechanismic account and issues of explanation and discovery. Therefore, we propose the notion of explanatory framework and, in advancing our account of the epistemic role of frameworks like the oncogene paradigm – in partial reformation of mechanismic explanatory monism – we aim at clarifying what can and what cannot be taken to be the role of such frameworks in the discovery of causal explanations.

The current mechanismic program can be seen as yet another reaction to the old hegemony of theoretical sciences as a model for science in general. While it might be true that theories are less prevalent in biology than, for instance, in physics (Keller 2004), this realization has, with the mechanismic account, led to a relative neglect of higher-level conceptualizations in epistemological analysis. For this reason, our account has points of contact with more traditional philosophical discussions surrounding scientific theories (see for instance the notion of *salience* in Kuhn 1992). However, our attempt is more specific: we propose an account intended to specifically trace the existing dynamics between overarching conceptions (explanatory frameworks) and mechanistic constructions. Furthermore, there are important differences between our account and traditional discussions of scientific theories. A first reason why we propose to use the expression "explanatory frameworks" referring to meta-mechanistic patterns of explanation is to avoid conflation between explanatory frameworks and theories as they are most often understood in philosophy of science, while at the same time preserving a space for the role of higher-level conceptualizations. In our view, explanatory frameworks sit at a higher level of generality and abstraction than mechanisms (and that is why it makes sense to speak of a meta-mechanistic level) but, importantly, they are not to be considered as theories in the classical sense. In the above-recalled family of uses referred to the oncogene explanatory framework, this linguistic conflation is instead just around the corner. Explanatory frameworks are indeed not theories in a very specific sense that, to be sure, is however clear in the mind of both historians and scientists who refer, albeit in a very loose sense, to the oncogene paradigm as a "set of theories". Two characteristics distinguish explanatory frameworks form theories. First, they do not state laws of nature and as a consequence are not conducive to law-like (Hempel 1965) or probabilistic (Railton 1978) generalizations and deductions, nor are they a family of models sharing semantic conditions (Thompson 1989). Indeed, explanatory frameworks are not amenable to falsification: rather than being in exclusive competition, they can co-exist or can be gradually displaced. As we will show, these changes and continuities differ considerably from the ruptures characterizing Kuhnian "paradigm shifts".

Another important characteristic of explanatory frameworks is that they incorporate a variety of elements (which may at least in principle account for the variety of designations recalled above). An explanatory framework – like in the case of the oncogene paradigm – is made of causal patterns, schemata (in the sense of Darden 2002), intuitions, hypotheses, evidential standards and

various bits of evidence and data coming from different experimental settings and instrumental devices. In our view, this enlarged family of inscriptions and material practices (Rheinberger 1997) is not itself the *locus* of biological explanation (in other words, it is not the *explanans*). We agree with the mechanismic account as to the fact that explanations in the life sciences generally come in the form of mechanisms. However, we are not convinced that an account of the discovery and provision of mechanistic explanations can be limited to the level of mechanisms. Our claims concern both the discovery of mechanisms and their explanatory usage. Scientific change is not just a piecemeal refinement of previously conceived schemata (Bechtel and Abrahamsen 2005; Darden 2002, see *infra* section 8 for a more specific discussion of this topic). Moreover, if to explain is to offer a mechanism at the appropriate level of detail, the mechanismic account falls short of accounting for this appropriateness; it leaves open the question of how scientists decide on the appropriate level of granularity. Finally, if to provide an explanation requires giving "all of the relevant components" and "none of the irrelevant components" (Craver 2007, 140), mechanicism fails to account for how it is that scientists *actually* decide what, in the indefinitely ample web of entities and activities that make up an organism and cause a given phenotype, should acquire explanatory relevance in the relatively narrow descriptions that mechanisms allow for.

So far, we have only provided a negative definition of what explanatory frameworks are, that is, we have shown in what ways they differ from what we have called mechanismic accounts of explanation. On the positive side, instead, we argue that explanatory frameworks account for the role of meta-mechanistic conceptualizations in guiding: a) the discovery and construction of mechanisms and b) their placement under specific explanatory lens. In this respect, they provide an answer to the problematic issues faced by mechanismic accounts as we have highlighted so far. To precise this claim and provide it with empirical support, we show in the next section (4) how explanatory mechanisms perform the two functions (a-b) outlined above in the case of the mechanistic characterization of TS gene p53 in the transition from the oncogene to the cancer-stem-cell explanatory framework. The reconstruction will substantiate the claims made so far and, finally, lead to a better characterization of explanatory frameworks in section 5.

4. Explanatory frameworks at work

In this section we provide evidence for the two main features of explanatory frameworks recalled above by elaborating on an existing body of scientific literature. We will describe two succeeding explanatory frameworks that have been (and still are) of major importance in molecular oncology. However, in order to avoid heavier locutions, we will use the most common wordings for these explanatory frameworks, namely the "oncogene paradigm" and the "cancer stem cell hypothesis". Nevertheless, it should be clear that we consider them both as full-fledged explanatory frameworks.

4.1. P53 and the oncogene paradigm

P53 is now one of the best-characterized genes in molecular oncology. It has been known to be involved in cancer ever since pioneering experiments on simian virus 40 (SV40)-induced tumors in the Seventies. Protein p53, barely detectable in normal cells, was observed to abundantly co-immunoprecipitate with SV40 T-antigen in both SV40-infected cells and in murine embryonal caricinoma cells. However, the meaning of this phenomenon changed substantially since this discovery. Today, p53 mutations are among the most notorious causes of malignant transformation. At the time, however, its potential causal agency in carcinogenesis was not even hypothesized. It was rather hypothesized that the oncoviral infection (or the malignant transformation itself) could trigger its production (Linzer and Levine 1979). In other words, p53 over-expression was mainly considered a by-product of viral transformation, rather than a causing agent.

To be sure, evidence for this hypothesis was not there, but the dominant understanding of cancer in that period stressed the fact that cancer resulted from a deregulation – what Morange (1997) calls the "regulatory vision" of cancer – which came to be expressed in transcriptional terms (Pitot and Heidelberger 1963). This deregulation was thought to be most likely the result of a viral infection. As the NIH Virus Cancer Program (1964-1978) shows, the belief that cancer was mostly caused by a virus was an enduring one. This view was dominant notwithstanding the fact that radiation had long been known to be mutagenic (Müller 1930), and that experimentation on radiation had shown that it could cause cancer (see for instance the BEAR committee, 1954-1964). These facts, however, were all interpreted within the regulatory vision of cancer, and therefore, although they were not ignored, their causal relevance was marginalized. While p53 over-expression might have been deemed causally relevant to transformation in the regulatory vision (as one of the gene deregulated by the viral infection), its role could at best be seen as a downstream intermediate to the transformative event.

As Morange (1993; 1997) showed, discoveries such as the realization that known carcinogens were also mutagens (McCann et al. 1975) and that transforming viral genes could also be present in uninfected cells (Stehelin et al. 1976) slowly fuelled the idea that cancer was, first and foremost, a mutation-driven genetic disease. This realization, in turn, eventually gave rise to a more successful explanatory framework: the oncogene paradigm. Although historians disagree as to the precise reasons for the success of the OG paradigm (see Fujimura 1988; 1992; Morange 1997), there is no doubt that in the decade from 1975 to 1985 this explanatory framework progressively gained prominence and steered molecular oncology in new directions. Therefore, some years after the initial observation of p53 over-expression in virus-induced tumors, and only within a new explanatory framework, p53 started to be studied as a potentially autonomous cause of cancer. As recalled above, in the early Eighties, p53 was already known to be *associated* with cancer because of its accumulation in transformed and embryonic cells (Mora, Chandrasekaran and McFarland 1980). From an experimental point of view, however, the elucidation of its *causal importance* in cancer owes to the availability of a screening technique that evolved within the OG paradigm. Let us quote directly from one of the papers[6] that, in 1984 established a causal role for p53 in cancer:

> "The previously observed cooperation of *ras* and *myc* oncogenes suggests an experimental test by which the oncogenic function of other genes can be assayed: a DNA clone carrying a gene of interest is co-transfected with either a *ras* or a *myc* clone and the appearance of REF [rat embryo fibroblasts] foci then scored" (Parada et al. 1984, 650)

The co-transfection experiment of rat embryo fibroblasts with two distinct plasmids, one encoding for ras and the other for p53, provoked malignant transformation of the cells, a phenotype that nor p53, nor ras alone were able to confer, thereby demonstrating a causative role also for p53. Due to this capacity, the authors realized that "the expression of p53 can be more than a consequence of the cancer state" (Parada et al. 1984, 650) – as previously believed. Thanks to the emerging OG explanatory framework, and to its related conceptual and experimental tools, p53 could now be recognized as a genetic *cause* of cancer, that is to say, as an *oncogene*.

This is just the first part of the story we want to tell about p53 and its molecular characterization, but let us pause here for a moment. We would like to stress that, in what we said so far, it is the experimental apparatus embedded in the OG paradigm that enabled the classification of p53 as an oncogene. In our terminology hence, the subsequent mechanistic explanations of the p53 network (more on this in the next section) in carcinogenesis and tumor development were made possible by the OG explanatory framework. Before, scientists just regarded p53 as an epiphenomenon of cancer transformation, and did not include its activity as relevant in the explanatory mechanism of carcinogenesis. This point is crucial to stress. While the regulatory

6 Two other papers one on the same issue of *Nature* obtained similar results (Eliyahu et al. 1984; Jenkins et al. 1984)

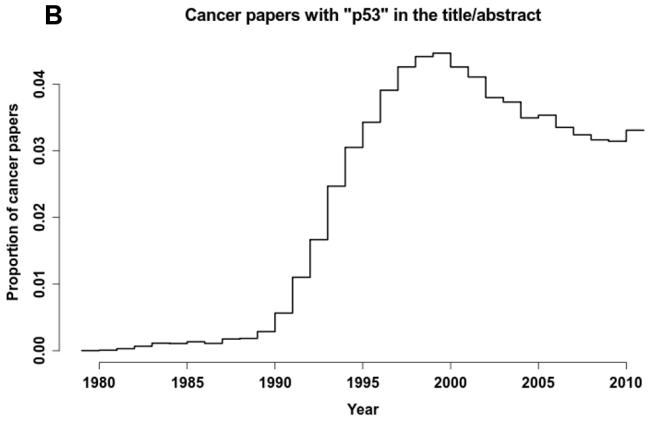
paradigm for the explanation of cancer, and its emphasis on viral transformation, dominated the scene, it simply made no sense for scientists to look for the mechanistic involvement of p53 in cancer. With the advent of the new OG paradigm instead, scientists had a reason – and an experimental tool – to check for the oncogenic nature of all the (then few) genes that were known to be associated with cancer, and p53 was one of them. It is the new paradigm that gives relevance to the p53 gene and drives the effort of a number of experimentalists to clarify its nature. This increased interest for p53 as an oncogene can also be confirmed by analysing the publication trends relative to the gene in the period following its association with cancer (table 1). The rate of publications per year referencing to "p53" in the title or in the abstract, normalized to the publication rate on cancer, increases by almost 5 folds immediately after p53 is classified as an oncogene with respect to the period following its generic involvement in cancer under the regulatory view.

A

Period	p53 articles	Cancer articles	Proportion of p53 articles	Increment relative to previous period
1979-1983	41	166,288	0.02%	-
1984-1987	196	167,189	0.12%	4.8 fold
1988-1991	616	201,634	0.31%	2.6 fold
1992-2011	54,184	1,592,882	3.40%	11.1 fold

Table 1. A. Result counts of Pubmed search for articles with "p53" in the title and/or abstract, relative to publications about cancer (loosely defined as "neoplasm" or "cancer" in any field) during the given period. 1984: p53 is classified as an oncogene; 1988: p53 is classified as a tumour suppressor gene; 1992: the first p53 knock-out (p53-/-) mouse is produced. **B.** Same results, plotted by year (non-cumulative).

B



The story of the classification of p53 as an oncogene and our data about publication trends show that it took the drive of the OG paradigm to start looking at p53 as relevant in causing cancer. This is particularly important for our account of mechanistic explanation in the life science. As our case demonstrates, the tortuous route towards the provision of molecular explanations is not accounted for by the sheer necessity to fill in the missing causal links in a mechanism. This is evident since, in the period between 1979 and 1984, the mechanism underlying p53 activity was not, and could hardly have been the focus of intense experimental research in the absence of any clue about the causal importance of this kind of mechanism for carcinogenesis. Instead, in the subsequent decade, as we show in the coming section, the OG paradigm and its associated experimental arsenal drove the detailed molecular characterization of a biological mechanism for p53 activity in cancer. While p53 over-expression was not new, the change in explanatory framework moved it from the margin to the centre of the attention.

Our reconstruction shows that the new explanatory framework steered research in new directions, but also that each explanatory framework reinterpreted previous knowledge and appraised its relevance in a new light. As Morange (1997) notes, for instance, viruses could also have a role to play in the oncogene paradigm, but this role was revised in light of the new explanatory framework (as carriers of oncogenes), and their relative importance in the general phenomena of cancer was gradually diminished. Today, viral infection is deemed relevant only to a minority of cancers. Likewise, the fact that many early oncogenes were involved in differentiation and development (Morange 1997), which was dear to the regulatory vision of cancer, was not strictly speaking falsified, but rather set aside and considered unimportant to an understanding of cancer. But it remained there, ready to be eventually recruited under other explanatory frameworks (see section 4.4).

4.2. P53: from oncogene to tumor suppressor

The pioneering experiments described above, that resulted in the classification of p53 as an oncogene, spurred a remarkable effort towards the characterization of the mechanism of action of p53, but soon turned out to be technically biased. In 1988 two groups reported that they were unable to replicate the co-transfection experiments, as, in their hands, some clones of the p53 gene did not complement ras in transforming normal cells into cancerous ones (for more details, see Lane and Benchimol 1990). Subsequent experiments soon revealed that the oncogenic features of p53 were indeed characteristic of a mutant form of the gene (fortuitously cloned in the available plasmids) that enhanced the production of the p53 protein well beyond the basal level of the non-transforming wild-type form of the gene (Eliyahu et al. 1988; Finlay et al. 1988; Hinds, Finlay and Levine 1989). The further elucidation of the oncogenic potential of p53 mutants led to a series of ground-breaking co-transfection experiments. Upon triple co-transfection of ras, mutated p53 and wild-type p53, Levine's group found that transfected cells did not acquire a malignant phenotype. The wild-type form of the gene was therefore able to exert a protective role against malignant transformation (Cathy A. Finlay, Hinds, and Levine 1989). This finding led to a re-classification of p53 "as a suppressor of transformation" (ivi) – in other words, a tumor suppressor gene (TS).

What remained to be clarified about the causative role of p53 was the reason why an activating mutation on a protective gene, instead of a repressive one, would drive the cell into malignant transformation. The answer came a few years later, when a dominant negative mutation[7] activity (Herskowitz 1987) of the p53 mutant was first postulated (Lane and Benchimol 1990) and eventually studied and characterized more deeply (Srivastava et al. 1993; Willis et al. 2004). With

7 A dominant negative mutation affects only one copy (allele) of the gene in question. The protein produced by the mutated allele, however, exerts an antagonistic activity on the wild-type (non mutated) allele of the same gene.

this realization the OG explanatory framework could finally deploy all its potential and direct in a specific way the further mechanistic characterization of the p53 pathway. The rate of p53-related publications per year literally exploded after the revelation of the dominant negative TS-nature of the gene and became remarkable as soon as the first knock-out ($p53^{-/-}$) mice became available in 1992 (Donehower et al. 1992). The plot above shows the increase in the proportion of p53-related publications in the decade following the production of the mouse model, and the sustained growth of the experimental work on p53 up to present days, when a total of more than 56,000 scientific paper is available on p53.

In other words, once the first dominant negative tumor suppressor gene was discovered, the OG paradigm could be extended to encompass yet another mechanisms of action. Again, this is very important for our account of the role of explanatory frameworks in the provision of mechanistic explanations. Before the recognition of the dominant negative-TS nature of the p53 gene, and thus before this further refinement of the OG paradigm, the mechanistic characterization of cancer could hardly be thought to bear on the protective capacity of genes acting in this way. Therefore, it is again the explanatory framework that drives the subsequent characterization of mechanisms, by establishing criteria of causal relevance for the phenomenon under investigation.

P53 is now known to be fundamental in response to DNA damage or replicative stress through its extensive involvement in cell cycle control, DNA repair, apoptosis, and permanent cell cycle arrest (i.e. cellular senescence). Due to its remarkable importance in the regulation of cell proliferation, TS p53 has been dubbed “the guardian of the genome” (Lane 1992). The mechanisms of action of p53 in different human tumors are now extensively characterized, and they give rise to an extremely complex network of activities and entities (see Vogelstein, Lane, and Levine 2000 for a general review). As we have tried to show, however, the causal relevance of p53 in cancer had first to be established, well before its mechanistic characterization could even be imagined. The OG explanatory framework, the concepts it contained and the experimental tools that were characteristic of its early years thus played a fundamental role in priming the direction of research towards a gene that did not, in and of itself, attract much attention in the context of the previous explanatory framework. The piecemeal mechanistic characterization of p53 is therefore the endpoint of a much broader current of ideas about carcinogenesis and related experimental techniques to probe them *in vitro* and *in vivo*. It follows that schema instantiation (Darden 2002), where it applies, is but a small part of the process by which mechanistic explanations are provided and explored. We provided evidence for this, drawing from both original scientific literature and from publication trend

analysis. We think that, instead, the mechanismic program has so far overlooked this feature of the life sciences by focussing exclusively on mechanisms as the privileged route to understand explanation and scientific change in molecular biology.

4.3. Cancer and stem cells

So far we have shown that explanatory frameworks orient mechanistic explanations towards causally relevant aspects of biological phenomena. In this section, alongside providing further evidence for this claim, we illustrate the second major characteristic of explanatory frameworks. As we said above, we take explanatory frameworks to be dynamic epistemic tools, in the sense that, by weighting differently the causal relevance of mechanistic explanations, they account for explanatory transitions in science. Our point here is to show that they so do without necessarily contradicting previously existing explanatory frameworks. In what follows, we substantiate this idea through a case study: the still on-going emergence of the cancer stem cell hypothesis and the role of p53 in the early mechanistic characterization of cancer stem cell biology in solid tumors.

The connection of cancer with development and stem cell biology is not a recent one in molecular biology. Cancer cells had been known to have embryo-like characteristics ever since pioneering studies on embryonal carcinoma cells and teratoma (see Andrews 2002 for an historical review). In particular, teratoma outgrowths containing a variety of mature tissues derived from all the three germ layers were soon described as disorganized embryos. Moreover, high levels of p53 were initially detected in cell lines of embryonic origin (embryonal carcinoma cell lines) in the early years of biochemical description of p53 precipitates. Therefore, the association between cancer, embryonic development and stem cells had been around for a long time before the experimental isolation of cancer initiating cells with stem cell-like characteristics. This major breakthrough in cancer biology took place only quite recently when, in 1997, John Dick and Dominique Bonnet demonstrated for the first time that human acute myeloid leukaemia (AML) has a hierarchical organization originating from uniquely tumorigenic blood cancer stem cell (Bonnet and Dick 1997). They attained this result through the convergence of a number of observations and experimental techniques. First of all, the blood compartment had long been known to be the result of a hierarchy of differentiated cells originating from long-term hematopoietic stem cells; and leukaemia had been observed to result in increased proliferation of progenitors. Therefore, it was natural to suspect that any of the cell types sitting higher in this hierarchy could have been the target of malignant transformation. Furthermore, efficacious experimental strategies for the immuno-phenotypical characterization (and the emerging CD nomenclature) of normal

hematopoietic stem cells were readily available[8] to oncologists to look at the stem cell-like properties of blood cancer cells. Bonnet and Dick thus applied those techniques and managed to isolate cancer cell possessing the same immune-phenotypic signature as normal hematopoietic stem cells. Upon transplantation into immune-compromised (NOD/SCID) mice, those cancer cells gave rise to leukaemia, therefore demonstrating their cancer-initiating potential. Furthermore, the leukaemia-initiating cells were able to self-renew upon serial transplantation and to differentiate in more mature cell types, thereby exhibiting the hallmark biological characteristics of stem cells.

The cancer stem cell (CSC) hypothesis, however, was at those times restricted to the blood compartment, whereas the application of this explanatory framework to solid tumors still lagged behind for a few years. At any rate a new explanatory framework was on the rise, and with it the program to apply it to other types of cancer. Breast cancer was the first solid tumor to be looked at as a stem cell disorder. In a famous PNAS paper published in April 2003, Hajj and colleagues adopted the cancer stem cell framework, originally advanced to account for acute myeloid leukaemia, to the case of breast cancer. They started from the observation that cells from solid tumors are considerably less competent in both forming colonies *in vitro* and in giving rise to new tumors *in vivo* in xenograft recipients (two assays that measure cells' tumorigenic potential). The only available explanations for these commonly observable features at that time were the so-called stochastic model of transformation – saying that tumors are made of pretty homogeneous cells all of which can transform, even though the necessary steps for transformation are "governed by low probability stochastic events" (Dick 2003, 3547) – and the so-called hierarchy model – that is to say the idea that, in line with Bonnet and Dick's findings on AML-initiating cells, cancer is due to very rare subsets of cell populations that possess unique tumorigenic potential. In the PNAS paper, the authors verified the second idea, and thus extended the cancer stem cell hypothesis to solid tumors for the first time. Again, their reasoning was driven both by the cognitive content of the CSC explanatory framework and by its toolbox of laboratory techniques: in brief, the isolation of breast CSCs was attained through immuno-phenotyping with CD markers and FACS-activated cell sorting – the same techniques used with AML a few years earlier, and the same technique that will be used to isolate brain cancer stem cells in another landmark paper published few months later, at the end of 2003, in *Cancer Research* (Singh et al. 2003). Interestingly, the PNAS paper was accompanied by an editorial written by John Dick who stressed that "purification of solid tumor T-IC [tumor-initiating cells] has been difficult because of the paucity of cell surface markers that enable cell sorting", and because T-IC xenograft assays from primary solid tumors were being mainly

8 For the history of the so-called monoclonal antibody revolution and CD-staining, and their role at the interface between oncology and developmental biology, see the contributions by Cambrosio and Keating (Cambrosio and Keating 1995; Cambrosio and Keating 2003).

conducted in *nude* mice, whose immune system is less compromised than that of the NOD/SCID mice used in the 2003 paper, and that could thus turn out to be rather difficult hosts for xenografts from human tumors. These comments highlight the importance of framework-specific experimental tools in the identification of causally relevant phenomena that are worth mechanistic characterization[9].

4.4. The emergence of a new explanatory framework: Cancer stem cells and p53

The emergence of the CSC hypothesis for the causal explanation of cancer gives us the opportunity to highlight a major feature of explanatory frameworks. As a new explanatory framework is introduced and gains attention in the scientific community, it is not necessarily the case that previous explanatory frameworks have to fade away or be falsified. In this section we bring evidence in favour of this feature by showing how recent findings as to the stem cell-like behaviour of cancer actually incorporate the oncogene paradigm. In the case we present below, p53 is attributed new causal roles within the new explanatory framework without invalidating the previous attributions of causal importance that, as we saw, led to the extensive mechanistic characterization of this gene within the oncogene paradigm. Therefore, the adoption of a new explanatory framework ought not be seen as the replacement of a theory by another, nor as a paradigm shift (Kuhn 1962): far from living "in another world", proponents of a new explanatory framework establish a continuity with previous conceptions by putting them into a new light[10]. This interactive co-existence shows that the turnover of explanatory frameworks does not take the form of ruptures between incommensurable paradigms. The dynamics of scientific change that we wish to highlight as typical of explanatory frameworks rather operates within the broader paradigm of molecular biology. These points can be illustrated by recent developments in the history of the p53 gene.

In 2009, Pier Paolo di Fiore and Pier Giuseppe Pelicci's groups published in *Cell* a crucial paper for the cancer stem cell explanatory framework, shedding new light over the cellular mechanisms that control breast cancer stem cell-like behaviour (Cicalese et al. 2009). The authors show that the tumorigenic potential of breast cancer stem cells is due to an unbalance in the rate of symmetric and asymmetric cell division, favouring the former over the latter. This is responsible for the sustained proliferation of cancer stem cells and, as a consequence, for the aggressiveness and invasiveness of this tumor. They show that the altered regulation of cell division is due to TS gene

9 Elsewhere some of the authors (1 and 2) have argued that local experimental constraints also influence the choice of model organisms in molecular oncology thus continuously reframing the evidential standards as to what counts as an explanatorily relevant model of cancer phenotypes (Maugeri and Blasimme, 2011).

10 This also distinguishes explanatory frameworks from styles of reasoning, which are much more pervasive and have an important dimension of incommensurability (Fleck 1979 [1935]).

p53: in the absence of the protective activity of this gene, wild type mammary stem cells proliferate indefinitely through a series of cell divisions that are mainly symmetric. Highly proliferative breast cancer stem cells, on their part, have almost undetectable levels of p53 signalling, which may account for their sustained self-renewal ability. Upon restoration of p53 to basal levels, indeed, these cells lose most of their self-renewal ability. The authors show that this happens because p53 is able to favour asymmetric over symmetric cell division in breast cancer stem cells. The molecular mechanisms responsible for the effect of p53 in self-renewal are yet to be clarified, but this study attributes a new explanatory role to p53 without invalidating its previous characterization as a tumor suppressor. More importantly, it reinterprets the already established functions of p53 in light of stem cell biology.

What is relevant about this paper to our present discussion is that it shows the full integration of the CSC hypothesis and the oncogene paradigm; additionally, it shows the distinct contribution of the former to set new standards of causal relevance, thus enabling further mechanistic explanations that could not have been previously visible in the however detailed mechanistic description of the p53 network. Here we showed how the cancer stem cell hypothesis for breast cancer, far from falsifying the oncogene paradigm, continues the mechanistic characterization of a major player in the OG paradigm like p53 under new explanatory lens. Its involvement in cancer as a regulator of symmetric-asymmetric cell division, however, is a result of the new explanatory framework. Indeed, until recently research on p53 was conducted mainly on relatively differentiated cancer cells (Bonizzi et al. 2012); within the cancer stem cell framework, there was now a strong rationale for studying it in stem cells. This already extensively studied gene (recall over 56,000 papers on this gene since 1979, see figure 1) is now the object of a novel mechanistic characterization due to the fact that its causal relevance is changed under the new explanatory framework. Therefore this transition happens without contradiction of neither the OG paradigm or the previously characterized mechanisms of the p53 network.

In the beginning of 2012, some of the same authors published an opinion paper on the emerging role of p53 in stem cell biology. As a concluding piece of evidence of the non-falsifying but yet productive nature of explanatory frameworks in setting new criteria of causal relevance for biological phenomena, it is worth quoting directly from this paper. According to the authors, accumulating evidence suggests that

> "p53 carries out its tumor suppression role not only via well-accepted functions, such as cell cycle inhibition and induction of apoptosis or senescence […] but also by regulating [stem cell] homeostasis" (Bonizzi et al. 2012, 11).

Note, however, that the new functions are not simply grafted onto an existing mechanism of p53

activity. Under the new explanatory framework, stem cell homeostasis becomes mechanistically associated with p53, both in normal and in malignant cells. Rather than simply refining existing mechanisms, the authors are integrating complex biological functions together and re-evaluating their meaning and relevance for a broad class of phenomena, in the light of the new explanatory framework.

5. Discussion and concluding remarks

Let us recapitulate the main points we presented in this paper. Explanatory frameworks are epistemic tools for the provision of scientific explanations. Although they integrate with mechanisms, they do not coincide with them. They attain to a higher level of *generality* than mechanisms, meaning that, the various features of biologically complex phenomena can be given different causal emphasis under different explanatory frameworks. Explanatory frameworks are therefore overarching patterns of explanation that subsume a variety of mechanisms and a multiplicity of diverse data under a common gaze. They are fundamental to explanation both for cognitive reasons (having to do with grasping the meaning of fine-grained description of molecular phenomena) and for directing experimental practices towards explanatory relevant findings. Our emphasis on the conceptual side of research in the life sciences, however, should not translate into a neglect of more pragmatic considerations. In the case of cancer, as we have highlighted, the turnover of explanatory frameworks is not only driven by explanatory or theoretical purposes, but also triggered by observations and specific questions raised at the clinical level. In particular, the latest realizations about the stem cell-like properties of cancer cells originate from the, often disappointing, therapeutic results of established strategies to fight the disease. One of the promises of the cancer stem cell hypothesis is the possibility of elucidating, and eventually targeting, those rare sub-populations of cancer cells that, due to their stem cell-like characteristics, escape conventional radio- and chemo-therapy and are ultimately responsible for tumor relapse and metastasis, indeed the real killers of cancer patients (World Health Organization 2012). The previous explanation for these relapses was the development of resistance to therapy through clonal evolution[11]. Once more, the cancer stem cell hypothesis generally does not deny that this phenomenon occurs, nor that it can account for at least some of the relapses. Rather, it states that in general the stem-cell-like properties of some cancer cells are *more relevant* to the understanding and avoidance of relapses. Standard mechanismic accounts of explanation, we argue, are unable to render fully the complexity of factors that orient experimental design and scientific discovery, and select the relevant mechanisms in the provision of an explanation. Explanatory frameworks can

11 See Germain (2012) for a philosophical analysis.

instead provide new insight to the mechanismic philosopher to appreciate how scientific activity is driven towards mechanistic explanations. This implies that explanatory frameworks cannot be reduced to any of the existing explications of what it is that provides "guidance in mechanism discovery" (Darden 2002). In particular, explanatory frameworks are neither schema instantiations, nor instances of modular subassembly, or forward-backward chaining. Let us briefly spell this point out.

According to Darden, "schema instantiations begin with a highly abstract framework for a mechanism, a schema, that is then rendered less abstract during the process of instantiation" (Darden 2002, 359). Regardless the obvious circularity of this definition, Darden's idea is quite understandable (also from the examples that she uses to illustrate this point and that we cannot report here). Explanatory frameworks, however, are not schemata themselves, mainly because they provide experimentally-backed, observation-laden reasons to think that schemata – and related mechanisms – might have this or that form. In the case of schema instantiation, as described by Darden, it is not at all clear what is the starting point of the abstraction process that eventually leads to the production of the schema. In particular, since the abstract form of the schema is supposed to represent the final mechanisms, it seems natural to think that schemata are abstracted from mechanisms. Should this be the case, however, we would have a *petitio principii*, since mechanisms would be both the starting point and the endpoint of schema instantiation. This account is therefore unable to tell where exactly mechanisms (and schemata) may come from in the first place.

Another proxy to mechanism discovery is modular subassembly. With this notion, Darden refers to the discovery of a working sub-component of a mechanism that is eventually applied, for example reasoning by analogy, to another phenomenon to be explained. This kind of reasoning is certainly present in actual scientific practice. However, the notion of modular subassembly does not account for the reason why a given module is taken to have causal relevance in the explanation of a phenomenon. Explanatory frameworks are instead exactly the kind of epistemic tools providing reasons to believe that a given part of the causal structure of a phenomenon deserves mechanistic explanation.

The last form of guidance to mechanism discovery, according to Darden, is forward/backward chaining. This amounts to reasoning about one part of a mechanism on the basis of what is known about its other parts. Again, this may well happen in science. However, even forward/backward chaining is unable to account for the causal relevance of that part of the mechanism that is still to be characterized, or for the changing weight attributed to some parts of the mechanism. In the case of the p53 mechanistic network, for example, by simply looking at the

already (extensively) characterized part of it there is not much one can say about the necessity of looking for p53's involvement in stem cell biology.

It might be correct to say that scientists generally look for explanations of a mechanistic form. However, this is far from saying something about how scientists are brought to attribute explanatory relevance to one mechanism (or part thereof) and not to another. Darden's proposals seem attractive generalizations of mechanism refinement but, as we showed, they are flawed in many respects as illustrations of mechanism discovery[12]. We therefore maintain that explanatory frameworks are a necessary complement in this respect.

In conclusion, we believe a correct account of explanation in biology needs to understand the role that explanatory frameworks have in shaping the discovery of mechanisms and at selectively discriminating among them. We argue here that this process of discrimination between relevant and non-relevant mechanisms or parts thereof is not at all a blind trial and error attempt, nor simply amounts to "fill in missing causal links in mechanistic model" (Craver and Darden, 2001). Rather it is a process driven by unifying conceptualizations indicating at the outset what kind of mechanisms have to be sought and the relevant class of entities involved. Most of molecular oncology does not proceed with the sheer aim of discovering mechanisms, but rather of discovering the relevant ones. Here, we are not debunking the mechanismic program *tout court*, but we express some dissatisfaction for specific features of it and suggest routes for improvements. We are suggesting that one should avoid thinking *only* about mechanisms. Mechanisms have taken the primary role in the philosophical accounts of biological explanation. The success of this research program rests, at least to a certain degree, on its ability to capture into one single account, the epistemic variability that characterizes the field of life sciences while, at the same time, maintaining a descriptive attitude with respect to the actual practices of scientists in this field. We argued however that, although insightful, the mechanismic program falls short of providing a truly faithful account of the ways in which biology is actually after mechanisms. Moreover, it fails to account for the ways in which major explanatory patterns alternate in molecular biology. This is probably due to an excess of focus on the ontological features of biological mechanisms, and partially to a certain resistance to acknowledge the role of higher-level conceptualization in molecular biology. The examples we provided show that changes in explanatory frameworks allow the reinterpretation of previous mechanisms and the development of mechanistic explanations into directions that would not otherwise be explored. Scientific novelties generally come in the form of mechanisms, but the

12 Darden herself is aware of this risk when she says that "[w]hether or not scientists actually used these reasoning strategies in their discoveries of mechanisms, the strategies could have been used. The strategies are what I call "compiled hindsight," that is, hindsight that can be extracted from an analysis of historical cases". This *proviso* however, leads to think that Darden's account is affected by a retrospective bias that deprives it of both normative and descriptive bite.

way in which mechanisms are considered to be relevant depends, we showed, on higher-level conceptualizations about the phenomenon at stake and the interests we have in it. Furthermore, our proposal tries to render the dynamics of continuity and discontinuity in life sciences. It does so, we have argued, by attributing a role to explanatory frameworks that the traditional epistemology of mechanisms has so far overlooked.

References

Andrews P.W. (2002). From teratocarcinomas to embryonic stem cells. *Philosophical Transactions of the Royal Society of London - Series B: Biological Sciences*, 357, 405-417.

Bechtel W. (2006). *Discovering cell mechanisms: The creation of modern cell biology.* Cambridge:Cambridge University Press.

Bechtel W. & Abrahamsen A. (2005). Explanation: A mechanist alternative. *Studies in History and Philosophy of Biological and Biomedical Sciences*, 36, 421-441. doi: 10.1016/j.shpsc.2005.03.010

Bechtel W. & Richardson R.C. (1993). *Discovering complexity: Decomposition and localization as strategies in scientific research*. Cambridge:The MIT Press.

Bishop J.M. (1995). Cancer: the rise of the genetic paradigm. *Genes & development*, 9, 1309-15.

Bonizzi G., Cicalese A., Insinga A., Pelicci P.G. (2012). The emerging role of p53 in stem cells. *Trends in molecular medicine*, 18, 6-12. doi: 10.1016/j.molmed.2011.08.002

Bonnet D. & Dick J.E. (1997). Human acute myeloid leukaemia is organized as a hierarchy that originates from a primitive hematopoietic cell. *Nature Medicine*, 3(7), 730-7.

Breivik J. & Gaudernack G. (1999). Carcinogenesis and natural selection: a new perspective to the genetics and epigenetics of colorectal cancer. *Advances in cancer research*, 76, 187-212.

Cambrosio A. & Keating P. (1992). A Matter of FACS: Constituting Novel Entities in Immunology. *Medical Anthropology Quarterly*, 6, 362-384. doi: 10.1525/maq.1992.6.4.02a00040

Cambrosio A. & Keating P. (1995). *Exquisite Specificity: The Monoclonal Antibody Revolution*. New York:Oxford University Press.

Cambrosio A. & Keating P. (2003). *Biomedical Platforms: Realigning the Normal and the Pathological in Late-Twentieth-Century Medicine*. Cambridge:The MIT Press.

Cicalese A., Bonizzi G., Pasi C.E., et al. (2009) The tumor suppressor p53 regulates polarity of self-renewing divisions in mammary stem cells. *Cell*, 138, 1083-1095.

Craver C.F. (2007). *Explaining the brain: Mechanisms and the mosaic unity of neuroscience*. Oxford:Clarendon Press.

Darden L. (2002). Strategies for Discovering Mechanisms: Schema Instantiation, Modular Subassembly, Forward/Backward Chaining. *Philosophy of Science*, 69, 354-365.

Darden L. (2008). Thinking Again about Biological Mechanisms. *Philosophy of Science*, 75, 958-969. doi: 10.1086/594538

Darden L. & Craver C. (2002) Strategies in the interfield discovery of the mechanism of protein synthesis. *Studies in History and Philosophy of Biological and Biomedical Sciences*, 33, 1-28. doi: 10.1016/S1369-8486(01)00021-8

Dick J.E. (2003). Breast cancer stem cells revealed. *Proceedings of the National Academy of Sciences of the United States of America*, 100, 3547-3549.

Donehower L.A., Harvey M., Slagle B.L., et al. (1992). Mice deficient for p53 are developmentally normal but

susceptible to spontaneous tumours. *Nature*, 356, 215-221.

Donnenberg V.S. & Donnenberg A.D. (2005). Multiple drug resistance in cancer revisited: the cancer stem cell hypothesis. *Journal of clinical pharmacology*, 45, 872-7. doi: 10.1177/0091270005276905

Eliyahu D., Goldfinger N., Pinhasi-Kimhi O., et al. (1988). Meth A fibrosarcoma cells express two transforming mutant p53 species. *Oncogene*, 3, 313-321.

Eliyahu D., Raz A., Gruss P., et al. (1984). Participation of p53 cellular tumour antigen in transformation of normal embryonic cells. *Nature*, 312, 646-649.

Finlay C.A., Hinds P.W. & Levine A.J. (1989). The p53 proto-oncogene can act as a suppressor of transformation. *Cell*, 57, 1083-1093. doi: 10.1016/0092-8674(89)90045-7

Finlay C.A., Hinds P.W., Tan T.H., et al. (1988). Activating mutations for transformation by p53 produce a gene product that forms an hsc70-p53 complex with an altered half-life. *Molecular and Cellular Biology*, 8, 531-9.

Fleck L. (1979). *Genesis and Development of a Scientific Fact*, Chicago: Chicago University Press (translation of Entstehung und Entwicklung einer wissenschaftlichen Tatsache. Benno Schwabe und Co., 1935).

Fujimura J.H. (1992). Crafting Science: Standardization Packages, Boundary Objects, and "Translation". In A. Pickering (Ed.), *Science as Practice and Culture* (168-211). Chicago:University of Chicago Press.

Fujimura J.H. (1988). The Molecular Biological Bandwagon in Cancer Research: Where Social Worlds Meet. *Social Problems*, 35, 261-283. doi: 10.1525/sp.1988.35.3.03a00050

Germain P.-L. (2012). Cancer cells and adaptive explanations. *Biology & Philosophy,* online first. doi: 10.1007/s10539-012-9334-2

Glennan S. (2005). Modeling mechanisms. *Studies in History and Philosophy of Biological and Biomedical Sciences*, 36, 443-464. doi: 10.1016/j.shpsc.2005.03.011

Glennan S. (2002). Rethinking Mechanistic Explanation. *Philosophy of Science*, 69, S342-S353. doi: 10.1086/341857

Hanahan D. & Weinberg R.A. (2000). The Hallmarks of Cancer. *Cell*, 100, 57-70.

Hempel C.G. (1965). *Aspects of Scientific Explanation: And Other Essays in the Philosophy of Science*. The Free Press.

Herskowitz I. (1987). Functional inactivation of genes by dominant negative mutations. *Nature*, 329, 219-222.

Hinds P., Finlay C. & Levine A.J. (1989). Mutation is required to activate the p53 gene for cooperation with the ras oncogene and transformation. *Journal of Virology*, 63, 739-746.

Hitchcock C. (1995). Salmon on explanatory relevance. *Philosophy of Science*, 62, 304-320.

Jenkins J.R., Rudge K., Redmond S. & Wade-Evans A. (1984). Cloning and expression analysis of full length mouse cDNA sequences encoding the transformation associated protein p53. *Nucleic Acids Research*, 12, 5609-5626.

Keating P. & Cambrosio A. (2012). *Cancer on Trial: Oncology as a New Style of Practice*. Chicago:University of Chicago Press .

Keating P., Cambrosio A. & Mackenzie M. (1992). The tools of the discipline: standards, models, and measures in the affinity/avidity controversy in immunology. In Clark A.E., Fujimura J.H. (Eds.) *The right tools for the job: at work in the Twentieth century life sciences* (312-354). Princeton University Press.

Kuhn T.S. (1965). *The Structure of Scientific Revolutions*, 3rd ed. University of Chicago Press. doi: 10.1023/A:1006474128258 (First published 1962)

Lane D.P. (1992). p53, guardian of the genome. *Nature*, 358, 15-16.

Lane D.P & Benchimol S. (1990). p53: Oncogene or anti-oncogene? *Genes & Development*, 4, 1-8.

Lewis D. (1986). Causal Explanation. In *Philosophical Papers*, Vol. II. Oxford University Press, 214-240.

Linzer D.I. & Levine A.J. (1979). Characterization of a 54K dalton cellular SV40 tumor antigen present in SV40-transformed cells and uninfected embryonal carcinoma cells. *Cell*, 17, 43-52.

Machamer P., Darden L. & Craver C.F. (2000). Thinking about mechanisms. *Philosophy of Science*, 67, 1-25. doi: 10.1086/392759

Maugeri P. & Blasimme A. (2011). Humanised models of cancer in molecular medicine: the experimental control of disanalogy. *History and Philosophy of the Life Sciences*, 33, 603-622.

McCann J., Choi E., Yamasaki E. & Ames B.N. (1975). Detection of carcinogens as mutagens in the Salmonella/microsome test: assay of 300 chemicals. *Proceedings of the National Academy of Sciences of the United States of America*, 72, 5135-5139.

Mora P.T., Chandrasekaran K. & McFarland V.W. (1980). An embryo protein induced by SV40 virus transformation of mouse cells. *Nature*, 288, 722-724.

Morange M. (1993). The discovery of cellular oncogenes. *History and philosophy of the life sciences*, 15, 45-58.

Morange M. (1997). From the Regulatory Vision of Cancer to the Oncogene Paradigm, 1975-1985. *Journal of the History of Biology*, 30, 1-29.

Muller H.J. (1930). Radiation and genetics. *The American Naturalist*, 64, 220-251.

Nicholson D.J. (2012). The concept of mechanism in biology. *Studies in history and philosophy of biological and biomedical sciences*, 43, 152-63. doi: 10.1016/j.shpsc.2011.05.014

Parada L.F., Land H., Weinberg R.A., et al. (1984). Cooperation between gene encoding p53 tumour antigen and ras in cellular transformation. *Nature*, 312, 649-51.

Pitot H.C. & Heidelberger C. (1963). Metabolic regulatory circuits and carcinogenesis. *Cancer Research*, 23, 1694-1700.

Railton P. (1978). Deductive-Nomological Model of Probabilistic Explanation. *Philosophy of Science*, 45, 206-226.

Rheinberger H.-J. (1997). *Toward a History of Epistemic Things: Synthesizing Proteins in the Test Tube*. Stanford:Stanford University Press

Salmon W.C. (1989). *Four Decades of Scientific Explanation*. Minneapolis:University of Minnesota Press.

Salmon W.C. (1984). *Scientific Explanation and the Causal Structure of the World*. Princetone: Princeton University Press.

Singh S.K., Clarke I.D., Terasaki M., et al. (2003). Identification of a cancer stem cell in human brain tumors. *Cancer Research*, 63, 5821-8. doi: 10.1038/nature03128

Skipper R.A. & Millstein R.L. (2005). Thinking about evolutionary mechanisms: natural selection. *Studies in history and philosophy of biological and biomedical sciences*, 36, 327-347.

Sonnenschein C. & Soto A.M. (2008). Theories of carcinogenesis: an emerging perspective. *Seminars in cancer biology*, 18, 372-377. doi: 10.1016/j.semcancer.2008.03.012.

Srivastava S., Wang S., Tong Y.A., et al. (1993). Dominant negative effect of a germ-line mutant p53: a step fostering tumorigenesis. *Cancer research*, 53, 4452-4455.

Stehelin D., Varmus H.E., Bishop J.M. & Vogt P.K. (1976). DNA related to the transforming gene(s) of avian sarcoma viruses is present in normal avian DNA. *Nature*, 260, 170-173.

Temin H.M. (1974). On the origin of the genes for neoplasia: G.H.A. Clowes memorial lecture. *Cancer Research*, 34(11), 2835-2841.

Thompson, P. (1989). *The Structure of Biological Theories*, Albany:State University of New York Press.

Vogelstein B. & Kinzler K.W. (1993). The multistep nature of cancer. *Trends in Genetics*, 9, 138-141.

Willis A., Jung E.J., Wakefield T. & Chen X. (2004). Mutant p53 exerts a dominant negative effect by preventing wild-type p53 from binding to the promoter of its target genes. *Oncogene*, 23, 2330-2338.

Wittgenstein L. (2001). *Philosophical Investigations*. Oxford: Blackwell Publishing (Translation of Philosophische Untersuchungen, 1953).

Woodward J. (1984). A theory of singular causal explanation. *Erkenntnis*, 21, 231-262. doi: 10.1007/BF00169275

Woodward J. (2003). *Making things happen: A theory of causal explanation*. New York:Oxford University Press.

World Health Organization (2012). *Cancer: fact sheet*. Geneva:World Health Organization. Available at: http://www.who.int/mediacentre/factsheets/fs297/en/ (last accessed: October 9th, 2012)