

Process and Practice: Understanding the Nature of Molecules

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Abstract: In recent years philosophers of biology have made renewed efforts to develop and defend a process ontology. These efforts have often focused on the example of living systems, which provide a strong case for a processual view of biological entities. Here I will analyze a different kind of biological entity, namely macromolecules. Looking at protein biology, I will show that contemporary theories in this field present us with a substance-like picture of macromolecules. Whilst this poses a challenge for existing process accounts, I will argue that the challenge can be overcome if metaphysicians abandon their focus on theory and follow a practice-informed scientific metaphysics. Turning to the practice of protein biology, and in particular the use of what I will refer to as ‘energy-level management’ practices, will suggest that macromolecules are processes, much like organisms.

Keywords: *process ontology, metaphysics, scientific practice, protein biology, intrinsically disordered proteins.*

1. Introduction

In recent years metaphysics has again become a central topic in philosophy of science. There are at least two strands to this new metaphysics. First, there have been renewed efforts to develop and defend a process ontology (see, e.g., Dupré 2012, Nicholson & Dupré 2018). Second, there have been efforts to promote specific ways of *doing* metaphysics. In particular the idea that metaphysics should be scientific metaphysics (*i.e.*, metaphysics that is informed and guided by the best available scientific knowledge) has gained a lot of traction (Hawley 2006, Ladyman *et al.* 2007, French & McKenzie 2012, Ross *et al.* 2013, Slater & Yudell 2017, Chakravartty 2017, Bryant 2020a, 2020b, Guay & Pradeu 2020, Sider 2020).

Interestingly, process philosophers could be seen as early adopters of scientific metaphysics. Whitehead’s process thought, for instance, was strongly inspired by developments in physics and the fundamental transformations

that happened in the field in the early 20th century. More recently, scholars arguing for process ontology have turned (again, one might add¹) to the life sciences to find motivation and support for their position (see, e.g., Dupré 2012, Bapteste & Dupré 2013, Ulanowicz 2013, Jaeger & Monk 2015, and individual contributions in Nicholson & Dupré 2018).

Of particular importance to this latter strand of process thought is the fact of metabolism. This complex set of chemical reactions and the exchange of material and energy that goes along with it are often seen as a key feature of living systems (Dupré & O'Malley 2009). Without the constant change that these processes entail, organisms would not exist. For living systems, a certain dynamicity is thus seen as fundamental, supporting the idea that they should be understood as processes, rather than as static things. As Nicholson & Dupré put it:

[T]he reality of metabolism forces us to recognise that organisms, despite their apparent fixity and solidity, are not material things but fluid processes [...]. As processes, and unlike things or substances, organisms have to undergo constant change to continue to be the entities that they are. [Nicholson & Dupré 2018, p. 17]

Importantly, such a process view of living systems not only emphasizes the importance of change and of what could be called 'dynamic stability' (Nicholson 2018). It also stresses the importance of interaction and hence relationality. 'Ex-change' with the environment is essential for the organism's identity (Meincke 2019) and at its core, metabolism is a fundamentally collaborative characteristic of living systems (Dupré & O'Malley 2009). For the contemporary process theories that are the focus of this paper, both dynamicity and relationality are therefore fundamental aspects of the world (see Section 2 for more details on this point).

However, the power of the example of living systems also raises the question of scope. If a key strength of the biology-inspired process views is based on the example of living systems, then what happens if we move our focus to other biological entities, such as macromolecules? These entities, which include protein or DNA molecules, are not metabolic systems. They are usually not seen as living beings that require a constant turnover of matter and energy in order to continue to be the entities they are.² Does the power of the organism-based arguments for process theory also transfer to these biological entities or does the example uncover their limitations?

This worry about scope is immediately deepened if we look at scientific models of macromolecules: whilst the theoretical models of living systems convincingly align with a process rather than a substance view (Meincke 2019), molecules are often presented by scientists as self-contained and self-sufficient entities that look more like substances than processes (see Section 3 for more details).

In this paper I will argue that this challenge from the macromolecular realm can be met by process philosophy. Macromolecules are as processual as living systems. But in order to convincingly make the case for this thesis, philosophers need to look not only at theoretical models in biology, but also at the details of scientific practice at the level of the laboratory bench. If we follow such a shift in philosophical methodology, it will be possible to show that whilst molecules appear to be substance-like, they are in fact processual entities that are made to look like substances. This is achieved through the use of a set of research practices, in particular of what I will refer to as ‘energy-level management’ (ELM) practices.³

In Section 2, I will say more about current process thought and the importance of dynamicity and relationality for these accounts. In Section 3, I will substantiate the claim that looking at theoretical models of macromolecules presents us with a strong substance-like picture of these entities. To make this point I will use the case of protein biology and the discovery of intrinsically disordered proteins. In Section 4, I will show that by switching the focus away from theoretical models and towards scientific practice, a different picture of molecules starts to emerge from contemporary science. This picture shows that whilst proteins might behave and look like stable things, this stability is produced and maintained by specific research practices, such as cooling or buffering. The need for such practices points us to the underlying dynamicity and relationality of proteins. In Section 5, I will conclude that by combining a focus on scientific practice and a theoretical analysis of the biological sciences, a strong case can be made for the processual nature of macromolecules.

2. Process Thought and Its Key Concepts

Process thought is an old position that can be traced back all the way to Heraclitus (Rescher 1996). Over the centuries, a number of process-based frameworks have been developed and there is a fundamental consensus among these different positions that being is dynamic rather than static. But at the same time there are also fundamental differences in the ways in which this basic assumption is transformed into different theoretical frameworks (Seibt 2020). The question of which of these competing frameworks holds the most promise has, as of yet, no clear answer.

One way of working around this issue is to treat process ontology not so much as a well-defined theory, but rather as a ‘point of view’ (Rescher 1996, p. 34). Such a point of view, Rescher argues, employs the concept of ‘process’ as a ‘categorical concept’, a thought-instrument that can be used to organize

our knowledge and experience of the world. Ultimately, what characterizes a process view are the different priorities it sets (always compared to its counterpart, a substance view) regarding key aspects of the world. The problem of arguing for a process or a substance view thus becomes the problem of arguing for how particular priorities are set.

To give an example of what Rescher has in mind here: whereas a substance view would, for instance, emphasize discrete individuality as a fundamental aspect of reality, process philosophers would stress the importance of ‘interactive relatedness’. Similarly, whereas a substance perspective would prioritize ‘fixity’ as a key feature of the entities that make up the world, a process view would stress change as a fundamental aspect of what exists. Other distinctions for which different priorities are set by a process and a substance view are separateness versus wholeness, or being versus becoming.

When we put these different priorities together, we might not end up with a precise definition of what a process is (or a substance for that matter). But we obtain a list of different aspects of reality that characterize a process world: relatedness, wholeness, becoming, change. These or similar properties are usually treated by process ontologists as fundamental aspects of the universe.⁴

A substance view, then, is a view that stresses ‘discrete individuality’ and ‘being’ (rather than becoming). In a substance ontology the world is treated as something that consists of ‘things’ – isolated and pre-existing entities that might (but don’t have to) engage in relations with each other. Interactions can take place, but they are secondary in the sense that they do not matter for the identity of the entities that interact with each other; a substance is what it is independent of whether or not relations occur. This is a view that can be described as a ‘disconnected boxes’ model of the world (Birch & Cobb 1981).

The starting point of the process philosopher is a different one. It is not just that they see the world as fundamentally dynamic. These dynamics, as highlighted in the introduction, are also intertwined with the importance of relations. Especially in the case of living systems, relations are seen as part of what makes the process the entity it is; they are part of what influences or ‘determines’ the nature of the entity or process. Such emphasis on the fundamental importance of relations is moving us away from a vision of disconnected boxes and towards a view of the world as fundamentally interconnected. This is also why many process philosophers have shown interest in how biologists describe the living systems they study, be it the growth, development, and maintenance of organisms or also more complex systems, such as termite colonies and their symbiotic nature (see, *e.g.*, Henning 2013, Guttinger 2018).

2.1 But what about molecules..?

What is interesting about the examples that current process philosophers use to illustrate, motivate, and/or support their position is that molecules are usually not their key focus. Of course, molecules will figure in them, for instance as the entities that are produced and/or consumed in metabolic reactions, or as the entities that are transferred between interacting organs or organisms. But the point of these examples is to say something about the fundamentally dynamic and relational nature of organisms and cells, not molecules.⁵

Molecules, however, not only represent a crucial part of any biological system (and have therefore become a key object of biological research over the last 80 years or so). They also represent a hard case for a process ontology because the molecular models that biologists work with, in contrast to the theories of organisms, leave little or no space for the fundamental importance of relations. As I will show in more detail in the next section, in theory molecules such as proteins look much like substances, *i.e.*, self-sufficient entities that *can* engage in interactions, but which ultimately have an unchanging core that is not affected by what is happening around them.

3. Protein Biology and the Role of Relations

Proteins play a prime role in the biological sciences, as they are seen as the entities that give structure to and drive almost every process that happens within a cell. The way proteins work is usually captured by the so-called sequence-structure-function (SSF) paradigm, a paradigm that has dominated protein biology for most of the 20th century.

3.1 The SSF paradigm and protein structure

The SSF paradigm consists of two key claims: (1) the three-dimensional structure (or ‘fold’) of a protein determines its function and (2) the fold itself is determined by the amino acid sequence (also called the ‘primary structure’ or ‘microstructure’ of the protein).

Biologists distinguish between two higher levels of protein structure, ‘secondary’ and ‘tertiary’ structure. The level of secondary structure is usually subdivided into key structural elements such as the ‘alpha helix’ or the ‘beta sheet’. These are relatively short motifs (often somewhere between 5 and 30 amino acids in length) that are connected by short linker sequences. These linkers are highly flexible and therefore allow the secondary structural ele-

ments to be rearranged into a more complex three-dimensional structure, which is called the ‘tertiary structure’ of a protein.

Importantly, the SSF paradigm postulates that this three-dimensional structure defines the function of the protein: because of their fold, proteins expose specific amino acids or patches of amino acids on their surface and it is these entities on the surface that give the protein the key features that allow it to interact with other proteins or molecules (for a review see Keskin *et al.* 2008). Many protein-protein interactions, for instance, depend on the presence of matching acidic/basic or hydrophobic patches on the surface of the different interacting proteins. Similarly, enzymes often display a structure (within a region called the ‘active site’) that matches the structure of their substrate(s) – if not exactly like a key fits a lock then at least like a glove fits a hand.⁶ According to the SSF paradigm, the relation between the molecular structure of the protein and its functional role(s) is linear and hierarchical: from the (lower) sequence level we get to the higher-order structure and ultimately the function of the protein.

3.2 The SSF paradigm and relations

Relations play a central role within the SSF paradigm, as proteins depend on interactions with their context (for instance the presence/absence of co-factors such as chaperones or magnesium ions) in order to fold properly or to execute their function(s). At the level of microstructure, however, relations do not figure in the SSF paradigm in any fundamental role. The microstructure is of course based on a specific relation between different amino acids (amino acid X comes before Y and after Z, *etc.*), but this sequence is treated as an intrinsic feature of proteins: a protein simply possesses a particular microstructure and interactions with other entities or processes outside of that structure are not part of what makes the microstructure the entity it is.

This limited role of relations in the traditional picture of proteins (and its hierarchical structure) is nicely captured by the philosopher Mark Goodwin (2011) who writes (in a paper on the classification of proteins):

The capacity to assume [a] tertiary structure (in the right biological circumstances) is [...] a consequence of the primary structure of the protein. So, although there are a lot of environmental and contextual factors to consider as well, the diverse functions of these sorts of [multi-functional] proteins are ultimately understood to issue, as potentials or capacities, from their primary structure.

Relations, in this picture, do not affect the character of the protein itself, which is fully defined by its microstructure. If we therefore follow how protein biologists and also philosophers *talk* about proteins (and other macro-

molecules for that matter) we get a picture of molecules as substances, *i.e.*, well-defined individuals for which relations are not fundamental.

3.3 A new processual understanding of proteins? The discovery of IDPs

The late 1990s were an important time of change in protein biology as new technologies, in particular the emergence of nuclear magnetic resonance (NMR) spectroscopy and small-angle X-ray scattering, brought about a shift in scientists' understanding of proteins. Whereas earlier the focus of protein biologists was firmly on structure and order, the new data suggested that constant structural change, or disorder, is fundamental for the functioning of proteins.

Once aware of the importance of disorder researchers quickly found that it is not a rare feature as up to 50% of proteins are now thought to contain disordered elements. This new class of proteins was labelled 'intrinsically disordered proteins' or 'IDPs' (Dunker *et al.* 2001, Dyson & Wright 2005, Uversky & Dunker 2010).

The discovery of IDPs clearly posed a challenge for the traditional picture of proteins as represented by the SSF paradigm. The interesting question in the context of this paper is whether this challenge was transformative, *i.e.* whether it changed theory in protein biology away from the substance-like picture we have encountered above to a more processual view of proteins.⁷

3.4 No room for change – understanding protein disorder

There are three different dimensions along which the disorder of IDPs can be characterized. The first of these dimensions is the *intensity* of disorder: as researchers are identifying more and more IDPs it becomes clear that there is a scale of disorder, ranging from complete disorder to what are called 'molten-globule-like' states. Complete disorder means that the polypeptide chain shows random-coil-like behavior, meaning that its amino acids move around using the maximum freedom of movement that the covalent bonds of the polypeptide allow for. 'Molten-globule-like' disorder represents a hybrid between a random-coil and a fully ordered state.⁸ An IDP that displays molten-globule-like disorder assumes on average a more compact shape than a randomly disordered IDP and might even contain some stable secondary structural elements. These IDPs are still highly dynamic entities (structurally speaking) but certainly less so than random-coil IDPs.

IDPs can also differ in the *extent* of disorder they display. As mentioned above, it is not always the whole polypeptide chain of an IDP that is disordered. The disorder can be localized to a short stretch (sometimes as short as 20-30 amino acids), a single domain, or the whole length of the protein. Be-

cause of these differences researchers distinguish between IDPs (which are taken to be disordered along the whole length of their polypeptide chain) and ‘intrinsically disordered regions’ (IDRs), *i.e.*, cases in which only a domain or a short stretch of the polypeptide display disorder (Dunker *et al.* 2013, Uversky 2013a).

The third dimension along which IDPs can be distinguished is the way in which disorder relates to the functional state of a protein. As for any protein, ‘to function’ usually means to interact with other molecules. For IDPs there are, roughly speaking, two options to do so: in many known cases the IDP/IDR folds into a specific (and stabilized) conformation upon binding to its interaction partner (which does not need to be another protein but can also be, for instance, a DNA or RNA molecule). This mode of functioning is referred to as ‘folding-upon-binding’ (for a review see Dyson & Wright 2002). Alternatively, some IDPs simply remain unfolded even when they are engaged in a functional complex with another factor.⁹

An interesting example of the first mode of functioning is p53, a key cell cycle regulator and tumor suppressor that contains an IDR in its N terminus (Wells *et al.* 2008). Researchers have shown that the IDR of p53 takes on different folds depending on which of its many binding partners it interacts with (for an overview see Uversky *et al.* 2009). Because of its disorder and folding-upon-binding behavior the IDR of p53 allows this crucial cell cycle regulator to take part in diverse complexes with very different functional consequences, explaining its involvement in a vast range of cellular processes.

An example of the second mode of functioning is Sic1, a yeast protein that is involved in the regulation of cell cycle progression (Deshaies & Ferrell 2001, Nash *et al.* 2001). Sic1 not only remains highly disordered even when it is interacting with its target protein, but the disorder also plays a key functional role as it creates an average electrostatic cloud around the protein that is required for its binding to its target factor (Borg *et al.* 2007, Mittag *et al.* 2008). Clearly, the need for constant change emerges as a central feature of this mode of functioning.

IDP researchers were acutely aware of the fact that the discovery of IDPs poses a significant challenge to the traditional SSF paradigm. It is therefore hardly surprising that they repeatedly called for a revision of the paradigm. Vladimir Uversky, one of the pioneers of IDP research, writes:

[T]he existence [of IDPs] questions one of the cornerstones in protein biology, chemistry and physics, that is, the structure–function paradigm. This concept claims that a specific function of a protein is determined by its unique and rigid three-dimensional (3D) structure. [Uversky 2002, p. 739]

Dunker and colleagues come to a similar conclusion:

[P]roteins with intrinsic disorder can in some cases carry out function without ever becoming ordered and thus remain disordered throughout their existence. To account for all of these possibilities, a new paradigm for protein structure/function is needed [...]. [Dunker *et al.* 2001, p. 50]

There is a strong sense here for a need to move to a new picture of proteins and protein function. The question is what this change in theory really amounts to, in particular from the perspective of a process ontologist. Is there a completely new picture of macromolecules emerging here, one that is built on ideas of dynamicity and relationality, rather than stability and autonomy? And if so, is there a general shift to a more processual understanding of the molecular realm taking place?

Ultimately, the answer to these questions has to be 'No'. In an intriguing way the main framework of the original SSF paradigm survived the challenge almost unscathed and managed to absorb the discovery of IDPs in a slightly amended form. There were at least two factors at play here. First, the challenge to the SSF paradigm that IDP researchers had in mind has had a limited scope from the very beginning: what these researchers were claiming is that there are *some* cases in which a fixed three-dimensional fold is not needed for a protein to be functional. Dunker and Obradovic (2001), for instance, propose that we need to think about protein function in terms of what they call the 'protein trinity', which consists of ordered proteins, molten-globules and random-coils. This already indicates that in their view the IDP discovery has led to a refinement of the SSF paradigm rather than a complete overthrow.

This limited scope of the challenge is important as it reflects a second point, namely the fact that IDP researchers do not challenge the strong focus on primary sequence that was a hallmark of the SSF paradigm. An obsession with microstructure remains a cornerstone of IDP research and the models it produces and works with. The overarching sequence-to-function link that the SSF paradigm is built on is not questioned by IDP researchers.

This is not something that immediately becomes clear when we read the early literature on IDPs, as these papers and reviews usually started with an emphasis on how new and important the discovery of IDPs is. But if we dig a bit deeper into the writings of IDP researchers it quickly becomes clear that they keep talking about microstructure as an intrinsic and fundamental property of proteins (including all forms of IDPs).

A good example of this entrenched way of talking can be found in an editorial for the journal *Intrinsically Disordered Proteins*, in which the editors (all key figures in IDP research) explain why they think that the term 'intrinsically disordered' is the best way of describing this new class of proteins (as opposed to other terms that were used in the past, such as 'vulnerable' or 'dancing' proteins). The editors state that the use of the term 'intrinsic' is warranted since "the lack of classical 3D structures represents an 'intrinsic' or

‘natural’ property of a protein, because it is encoded in its primary amino acid sequence” (Dunker *et al.* 2013, p. 2).

We find the same viewpoint expressed by Uversky (2013a, p. 700) who states that: “Each protein is believed to be a unique entity that has quite unique primary sequence [*sic*] which governs its 3D structure (or lack thereof) and ensures specific biological function(s).”

What is put in brackets in this second citation is crucial: Uversky, as well as the other researchers cited above, make the point that *both* order and disorder are encoded in the primary structure of the protein. The researchers do not intend to break with the basic assumptions that were also guiding the SSF paradigm, namely the idea (a) that the foundation of everything is provided by the microstructure of the protein and (b) that this microstructure can be treated as an intrinsic feature of proteins that does not rely on relations. What they work with is therefore nothing more than a slightly revised SSF paradigm, which could be referred to as the ‘sequence-function’ or ‘SF paradigm’ (as it maintains the overarching ‘sequence-to-function’ link that also characterized the original SSF paradigm).

All of this is relevant in the context of this paper. The IDP discovery has been revolutionary in many regards. But throughout this recent episode in protein biology, the researchers involved have not changed the way they represent the fundamental features of proteins. Protein biologists still represent proteins (and I would claim other molecules as well) as something that looks more like a substance than a process.

But what does this resilience of substance-like representations mean for the metaphysician? Does it mean that molecules simply *are* substances? If so then a process philosopher who takes scientific metaphysics seriously would have to conclude that her process framework does not apply to the (macro)molecular realm.

Whilst the above developments could be interpreted as a problem for the process philosopher, it is also important to remember the caveats that a number of philosophers have identified regarding the use of theory for metaphysical debate. Alan Love and Marco Nathan, for instance, have highlighted that the models that we encounter in science often contain a high level of abstraction and idealization (Love & Nathan 2015). In their analysis of mechanistic models, they show that in order to work with such models when doing metaphysics, philosophers also need to factor in scientists’ (explanatory) practice. As I will show in the next section, practice – and in particular laboratory-based research practice – also has to be factored in when assessing the nature of molecules.

4. Understanding Protein Microstructure: Theory and Practice

The analysis in Section 3 has shown an interesting view of microstructure that dominates the debate within IDP research (and protein biology more generally), namely the view that microstructure is a stable and non-relational feature of proteins. This take on microstructure, however, is at odds with other fields, such as protein chemistry, which tends to depict molecular structure as dynamic and relational. One way of illustrating this is by looking at the concept of the ‘protonation state’ of a protein.

4.1 Protein microstructure revisited

In protein chemistry the expression ‘protonation state’ is used to refer to the numbers of protons present in a protein. The number of protons present matters because they are positively charged entities. Any change in proton numbers will therefore directly affect those characteristics of the protein that are sensitive to changes in charge, such as its fold.

The protonation state of a protein can change because many amino acids contain protons they can give off to a proton acceptor (making them so-called ‘proton donors’) or because they have free electron pairs that can be used to bind an additional proton. As a consequence, protons are constantly exchanged between the protein and the molecules that surround it (primarily the bulk water molecules).

This exchange depends, among other things, on the pH of the surrounding solution: as the pH is nothing but a measure for the concentration of protons in a solution an increase or decrease of the pH means that more protons are pushed onto the protein or removed from it.

Because of this, the number of protons (and hence the atomic microstructure) of the protein can change whilst its amino acid sequence remains the same. Simply indicating the sequence of a protein does therefore not tell the researcher what its actual atomic microstructure is. To be able to do so the researcher will also have to know the parameters of the system the protein is part of. Relations therefore *are* entering the microstructural level even though the traditional SSF paradigm (and also the revised SF paradigm) in protein biology usually treat it as a non-relational and fundamental level.

It could be argued that all of this is merely an epistemic point that is fully compatible with a substance ontology: it might be the case that in order to *predict* the protonation state of a protein we need to know more than just its primary sequence. But such epistemic necessity would not mean that the world itself is processual. Protonation could simply be treated as the addition or removal of ‘things’ or substances.

Such an argument is part of a more general strategy that I will refer to here as the GOLD strategy (for ‘Go One Level Down’). The key move of this strategy is to acknowledge the dynamic nature of a higher-level phenomenon but to then go down one level (to single atoms in this case) and to claim that at this level we are no longer dealing with relational and dynamic entities. Whilst we might be forced to take relations into account at the higher level, this is – according to this strategy – merely an epistemic issue. Ontologically speaking nothing has changed, because the lower level is still composed of substances, *i.e.*, well-defined and stable things for which relations are merely secondary.

The GOLD strategy is a popular move, but it runs into problems when we start factoring in energy levels and related practices. To stick for a moment with the example of protons-as-substances: clearly, at the temperature and pressure levels used in protein biology (usually somewhere between 4 and 37 degrees Celsius and atmospheric pressure) a proton is a highly stable thing that for all intents and purposes can be treated like an autonomous, non-relational entity. But if we switch, for instance, to a context of high-energy physics where different forces and relations are brought into play, we will struggle to find this stable and autonomous entity. We enter a world in which the focus quickly shifts to ‘fields’ and complex assemblies of subatomic particles (which, it could be argued, are not particle-like at all). Physics – not just high-energy physics but also quantum physics – has unsurprisingly been a key influence on many process philosophers (Nicholas Rescher claimed that the rise of quantum theory “put money in the process philosopher’s bank account” (Rescher 1996, p. 97)).

It is this focus on energy and practice that can also help us to make sense of why microstructure has remained such a central part of the worldview of protein biologists. The IDP discovery meant that protein biologists had to allow for a highly dynamic and relational world at the level of protein structure and function. But with the GOLD strategy they could move down to the level of atomic microstructure and treat this as a stable and non-relational bedrock for their work. But as with the case of protons-as-substances, also in the case of the apparently stable microstructure this strategy only works if certain practices are put in place and if energy levels in the experimental system are managed.

If we turn our attention to the ways in which biologists handle proteins in everyday research, we realize the importance of ELM practices. These practices are so unassuming and widespread that they are easily overlooked. This can mislead the non-practitioner. The feature that matters most in the context of this paper is that these practices can be used to turn microstructure into a non-relational feature of proteins. ELM practices ultimately create a context in which the processual character of molecules becomes invisible and

can be safely ignored. This is also what allows researchers to work with the SSF/SF paradigm in the way they do and explains the resilience of the old picture of proteins as substance-like. It works because it is made to work, not because it is an accurate picture of what proteins are.

4.2 The importance of ELM

ELM practices come in many shapes and forms, from cooling down a solution to creating a vacuum in a test tube. They all have one goal: to control and manipulate the energy landscape in an experimental setup.

The key practice used to keep the protonation state of a protein stable is buffering. This term does not refer to what a computer is doing when streaming content online but to one of the greatest tools the experimental biologist has at her disposal, namely the use of buffered solutions.

A buffered solution is a specific mixture of an acid or a base and its corresponding salt, dissolved in water. What this solution can do is to absorb extra hydrogen ions that are added to the system (or to compensate for the addition of hydroxide ions (OH^-), which can trap free hydrogen ions). A buffer can thereby keep the pH stable even though protons are added to or removed from the solution.

Different acids and bases can be used to create different buffers. An often-used mixture to create an alkaline buffer ($\text{pH} > 7$) is a combination of ammonium chloride (NH_4Cl) and ammonium hydroxide (NH_4OH). Depending on how much of each substance is added to water the pH of this buffer can be adjusted to a range from pH 8 to pH 10 (each acid or base will have its specific range). If this buffer is set, for instance, to pH 9 it will remain at this level if protons are added (which would normally lead to a lowering of the pH).¹⁰

Apart from buffered solutions the practicing biologist will use other tools to control the stability of their protein of interest, such as tightly controlled temperature (protein solutions are usually handled on ice and stored at -20 or -80 degrees Celsius) or the salt concentration of the solution. All of these practices are part of ELM practices, as they are ultimately about the control of the energy landscape the protein is exposed to in the experimental setting.

4.3 Creating a non-relational world

Importantly, ELM practices are boundary-making practices: by setting a specific pH (and temperature *etc.*) the researcher fixes the protein in a specific state (in our example the protonation state). The protonation state of a protein is – among other things – a function of the pH. This function can be turned into a bijective function (meaning that for each pH there is a single corresponding protonation state) if all other parameters are kept constant

(again through ELM practices). If this is done and if the solution is buffered, then the protein will display a well-defined microstructure. In the context of ELM practices, the relational and dynamic nature of the protein microstructures therefore becomes invisible and its vague boundaries become sharp and fixed. The protein looks and behaves like a non-relational entity with a well-defined boundary.

Practicing scientists are of course acutely aware of the dynamic nature of proteins and the measures needed to keep them stable. This is why they use ELM practice in the first place and why they always specify not only the sequence of the protein but also the pH, the temperature, and the other elements of the system with which they are working.¹¹

When all of these measures are in place a space is created within which proteins can be treated and talked about as if they were substances. This is not some form of make-believe as the proteins actually behave like a well-defined and independent entity in this particular experimental setting. But this substance-like behavior depends on this constructed space, which covers up the fact that relations are a fundamental part of the entity of interest. This last point, however, only comes to the fore if we also focus on scientific practice when thinking about the nature of proteins. By just looking at the way protein biologists talk about or represent proteins the non-scientist would have a hard time recognizing this. Factoring in practice therefore serves as a discovery tool when reading the natural sciences and what they are telling us about the world. Much like Love & Nathan (2015) suggest in the context of mechanistic models, focusing on scientific practice helps uncover and emphasize the idealizations that are at work in laboratory-based science.

4.4 Flawed practices?

It could be argued that such a focus on practice is problematic because practices change all the time. And if philosophers want to draw metaphysical inferences by looking at practice, they should avoid being guided by practices that have turned out to be flawed and/or unreliable.

It is obvious that scientific practice is constantly evolving and that there are many new practices that are abandoned by scientists, simply because they did not work (or did not work reliably). However, the practices I consider here should not be understood as particular pieces of scientific technology that are linked to untested new materials or machines. The practices I consider here are processes such as cooling a specimen to slow down its decomposition. These are practices that have existed long before the rise of the modern experimental life sciences.

Researchers of course develop new ways of cooling objects, or they establish new buffer solutions that help them deal with particular practical chal-

lenges. But in all these different instantiations the base remains the same. ELM practices are not tightly bound to one particular way of realizing them or to one particular piece of machinery that could fail. Practices such as adjusting temperature or pressure have been proven to be powerful working principles again and again, giving them a stable status as established parts of scientific thinking. Taking these central parts of science into account when doing metaphysics therefore means to build on one of the backbones of the empirical sciences.

5. Conclusions

As I discussed in Section 2, many contemporary process frameworks take relationality to be a fundamental feature of the world. Systems such as organisms would not exist if it were not for their interactive relatedness. A ‘disconnected boxes’ view of the world thus cannot capture the nature of organisms.

A key problem for these process frameworks, however, is that molecules – as represented by scientists – do not seem to follow the example of organisms. As I have shown in Section 3, the main models used in protein biology do not assign a fundamental role to relations. A protein is the thing it is because of the intrinsic properties it carries (*e.g.*, microstructure), and not because of its relational nature. This non-relationist picture of proteins also does not change if we factor in recent developments, such as the discovery of IDPs, where dynamicity became a dominant focus of research. The molecular realm therefore poses an interesting challenge for contemporary process accounts.

But the example of protein biology has also allowed us to highlight the constructed nature of this state of affairs: proteins are made to look and behave like substances through the use of specific practices, in particular those that I have labelled ‘energy-level management’ practices.

Process philosophers such as Dupré (2012) propose that all ‘things’ should be understood as stabilized processes. Once we look at the natural sciences with a focus on its practices, we see that a key part of doing science is to actively make things stable and autonomous; solidifying phenomena is an integral part of scientific practice (Chang 2004, Feest 2011). This fact is easily overlooked if we only focus on the theories and models used in scientific explanations (what Waters (2017) calls the “traditional approach to scientific metaphysics”). For the process philosopher it is therefore instrumental to pursue a practice-informed scientific metaphysics, paying particular

attention to the ways in which researchers manipulate their objects of interest in the experimental context.

The point here is not to claim that a focus on practice *proves* that the world is processual. It is not the case that ELM practices tell us what the world looks like. They are not some sort of mirror of the world. The point of taking ELM practices into account is that this allows us to extract a different picture of molecules from the sciences, one that is without the idealizations that otherwise easily dominate and distort the debate. The move to practice is a move that changes our reading of what the natural sciences are presenting us with (Love & Nathan 2015). It is also a move that re-calibrates the outsider's eyes and ears. As part of this re-calibration, the fundamentally relational nature of molecules comes to the fore and the molecular realm starts to look less substance- and more process-like.

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Notes

- ¹ See Nicholson & Gawne 2015, Nicholson & Dupré 2018 for a discussion of how the so-called 'organicists' connected biology and process thought in the early 20th century.
- ² See Dupré & O'Malley 2009 for an interesting discussion of the boundaries of life in relation to macromolecules such as prions.
- ³ This shift in philosophical methodology follows the practice-focused approach to scientific metaphysics that has recently been advocated by (Waters 2017) and co-workers (<http://biological-practice-to-metaphysics.org>). It also aligns with a broader trend in contemporary philosophy of science, namely the focus on look-

ing at practice rather than just theory (see, e.g., Ankeny *et al.* 2011, Boumans & Leonelli 2013, and contributions in Soler *et al.* 2014).

- ⁴ This does not mean that separateness, being, or fixity do not matter in a processual universe. It just means that these are not treated as fundamental.
- ⁵ A rare exception is the enzymologist and process philosopher Ross Stein who has applied Birch and Cobb's process view to enzymes (Stein 2004, 2006). See Guttinger 2018 for a discussion of Stein's account.
- ⁶ The lock-and-key analogy refers to the famous model proposed by Fischer in 1894 (Fischer 1894). The hand-and-glove model is an adaption of Fischer's model, usually called the 'induced fit' model (Koshland 1958).
- ⁷ The discovery of IDPs, and the field of protein biology more generally, has given rise to an important set of philosophical research, in particular in the context of debates about natural kinds and classification (see, e.g., Slater 2009, Tobin 2010, Goodwin 2011, Bartol 2016, Havstad 2018, Tahko 2020).
- ⁸ The name 'molten-globule' was originally used by protein biologists to refer to an intermediate in the protein folding process and indicates a state in which the polypeptide has obtained its rough shape but in which not all amino acids have formed their final key interactions with other atoms within the polypeptide. In this intermediate state, the residues are not 'locked in', meaning they can still move freely to a significant degree (Ohgushi & Wada 1983, Arai & Kuwajima 2000).
- ⁹ There are also some cases in which a third mode of functioning is at work, namely an unfolding-upon-binding mode (Uversky 2013b). Here a transition from an ordered to an unordered state is required for the protein to fulfill its causal role(s).
- ¹⁰ This only applies to a certain degree as buffers can be exhausted if excess amounts of acid (or base) are added.
- ¹¹ These details will usually not appear in the main sections or figures of the paper but only in the Material & Methods part (and even there only in very cryptic form).

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