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The long life of unicorns*

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ABSTRACT

In this paper co-authored by a philosopher and a scientist, we aim to provide some philosophical and scientific insights into the long life of unicorns, such as how deep misconceptions can often persist, and some for a long time, in the scientific literature. We take as an example the belief, often seen in the bio-nano and nanomedicine literature, that nanoparticles have some special abilities at crossing lipid membranes.

KEYWORDS: Nanoparticles, Nanotechnology, Membrane, Uptake, Endocytosis, Diffusion, Evidence, Scientific Models, Theoretical Models, Philosophy, Peer Review

PURPOSE AND RATIONALE

In this paper co-authored by a philosopher and a scientist, we aim to provide some philosophical and scientific insights into the long life of unicorns, such as how deep misconceptions can often persist for a long time in the scientific literature. We first introduce our example of the "nanoparticle diffuse through membranes" unicorn, and then we use it to discuss three aspects that underpin the long life of unicorns: (1) the role of article introductions and references in sustaining the myth, (2) the role of models and theories in this context, and, finally (3) the impact of failures in rigorous experimental validation.

INTRODUCTION

The goal of science is to understand better physical reality, such as the real world around us, and not fictional creatures such as unicorns. Technology-oriented research, including much of nanotechnology, often aims not, or not solely, to understand phenomena, but also to produce objects. These objects can be devices, nanoparticles, or engineered surfaces with specific properties that enable either immediate or future applications. Thus, most articles in bio-nano science list a wide variety of applications as the rationale for the work. Those applications constitute entirely legitimate justifications if they do exist. However, they are sometimes like unicorns. They are mythical, and there is no realistic scenario where the objects produced in the article would lead to the proposed application. Arguably that does not constitute *studying unicorns* because while the envisioned applications may indeed be far-fetched, the nano-objects or devices they describe do exist. It may therefore be true to say that we might learn something useful by studying them. This counterargument does not entirely mitigate the problem of deceptive advertising of improbable applications to justify the research. There is a category of research articles even more akin to the study of unicorns than studies justified by unrealistic applications. That category is composed of articles where the phenomenon and/or the objects themselves, and not only their proposed applications, are mythical. Our focus in this perspective is on this latter category using modeling the passage of nanoparticles through lipid membranes as an example. There are plenty of unicorns in other areas of nanoscience, as humorously illustrated by the recent *ACS Nano* article[1] reporting on the performance of HU-GO-BD (Hummers Graphene Oxide Bird Droppings) for electrocatalytic applications.

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Figure 1: The Unicorns, "le bestiaire fantastique," 16th Century tapestry (Château de La Trémolière, France). Belief in the existence and curative properties of unicorns was common in the European Middle Age and Renaissance. The debate over their existence lasted well into the 18th century. (Wikipedia 2020[2])

On the definition and common usage of the term "nanoparticles"

Nanoparticles are objects made of any materials and which have at least one dimension smaller than 100 nm. In other words, *"nanoparticles" are not a class of materials but a size range of any material*. Thus, nano-size is one of many characteristics that would exist in any particular class of materials. Other properties include charge, hydrophobicity, magnetism, and others. While this definition is entirely reasonable, if "nanoparticles" was used purely in that sense, there would not be much to say about their properties or applications because *size range* does not have properties or applications. Yet, scientific articles, reviews, and scientific narratives destined for the broader public are full of promises and warnings about their properties and applications. Thus, it is commonly and maybe erroneously implied that there is something special about being "nano."

The "nanoparticles diffuse through membranes" unicorn

Thanks to their size or special structures, the concept of nanoparticles somehow magically getting into cells, thus causing toxicity or opening new ways to deliver drugs, is powerful. It has been effectively conveyed to the public's general understanding of science via hundreds of press releases stressing the benefits or the risks alternatively. However, the overwhelming evidence is that nanoparticles do not diffuse through cell membranes and that they enter cells by endocytosis. Gold colloids were already used as a tool (contrast agent) sixty years ago to study this biological phenomenon (Figure 2a).[3,4] The suggestion in 2005 of non-endocytotic transport of nanoparticles[5] was rapidly challenged (Figure 2b),[6] but the powerful and inspiring idea, for example, the unicorn, has lingered despite the lack of evidence. The fact that nanoparticles cannot easily enter cells is evidenced from a biological and evolutionary standpoint: cells have to protect themselves from viruses. This disappointing physical reality has not prevented the publication of a large number of theoretical articles studying the diffusion of nanoparticles through membranes as if that hypothetical phenomenon were relevant to drug delivery and nanoparticle toxicity.[7–13] We will argue that these articles, instead of helping the understanding of interactions between nanoparticles and cells, sustain the unicorn. For example, they add to the confusion over a simple scientific question, namely, "do nanoparticles enter cells by diffusing through lipid membranes?" which happens to have a simple answer, established for several decades: "No, they don't."

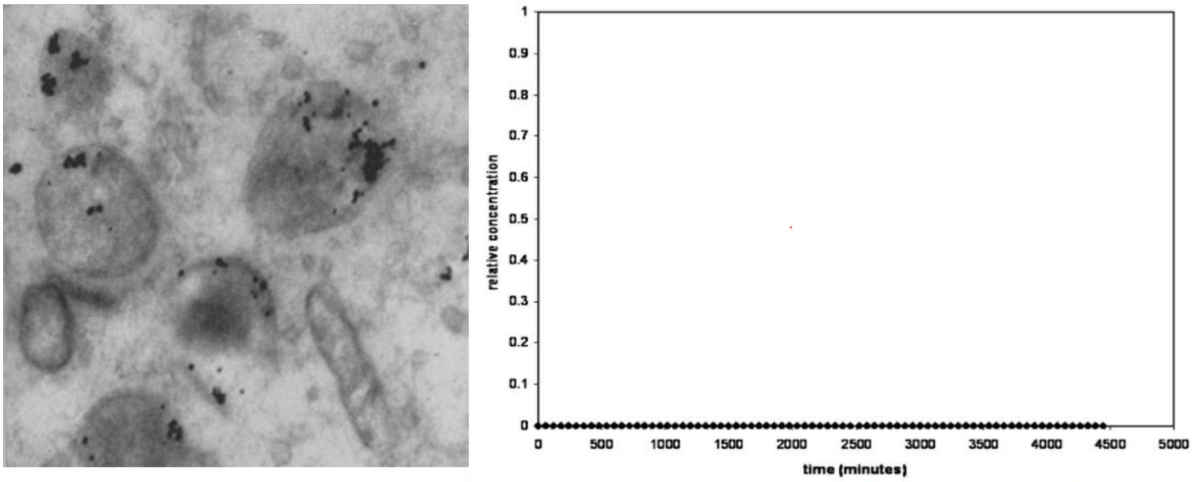

Figure 2. Gold nanoparticles enter cells by endocytosis and not by diffusion through membranes. (A) Gold nanoparticles in endosomes, Harford et al., 1957;[4] (B) Representative rate of transport of 7, 10 and 15 nm gold nanoparticles across lipid membranes; (Banerji and Hayes, 2007[6])

Critical discourse in scientific and philosophy journals

Demonstrating in sufficient detail how and where the articles studying unicorns go wrong requires a forensic dissection of the text and of the evidence. This is actively discouraged in scientific journals; [14–17] interestingly, the opposite is true in philosophy. Indeed, philosophy is built on the kind of analytical engagement that comes from critical discourse. In philosophy journals, ideas are challenged, rebuked, and even sometimes ridiculed. Most arguments in philosophy build on the structure, or the wreckage, provided by the arguments of another. As Priest notes, "philosophy is precisely that intellectual inquiry in which anything is open to critical challenge and scrutiny." [18] Without this capacity to challenge arguments and truth claims, the basis of philosophy would be compromised. This is not to say that the philosopher will always recognize their own biases, nor even that they will want to or even enjoy the process of being challenged. It's also worth noting that critical discourse without due care to the other can become highly personal and/or alienate those who might already occupy precarious outsider positions to the discipline.[19] What remains key, however, is that the principle of critical discourse remains at the heart of any sincere philosophical engagement. To the extent that a philosopher is capable of such, the impetus must be that any argument they make, or as made by any other philosopher, should withstand sustained critical and analytical scrutiny.

Laplane et al. suggest that applying philosophical methods to scientific endeavors might help remedy some of the weaknesses that can arise.[20] There is plenty to add regarding the benefits to philosophy that come from engagement with science. While we intend to discuss this point in a philosophy journal in due course, for the moment, our focus remains the unicorn. We surmise that the lack of open and public discussion of the evidence that supports scientific claims is one reason for the prosperity of unicorns. Here, new spaces for discussions such as Twitter, PubPeer, blogs, and online international journal clubs constitute interesting alternative avenues. It is not enough that scientific endeavors share many of the analytical tools found in philosophy if the disciplinary traditions of the former limit the scope for their application. These new critical spaces make it possible to apply logical and conceptual analysis to specific theoretical claims, which are often unwelcome in traditional scientific journals.[14–17]

How introductions sustain the unicorn?

Introductions of scientific articles are structured texts, which follow specific norms. They do more than introduce the topic; they justify the research and overview of the findings. How then, do authors build the case for studying unicorns in the introductions of their articles? Let us consider the case of the "nanoparticles-diffuse-through-membranes" unicorn again.

First, authors need to establish *plausibility*. They may, for example,[7] note that some small organic compounds *can* diffuse through lipid membranes. This is correct, although there is an increasing body of evidence that even for small molecular weight organic compounds, intracellular uptake occurs via protein transporters, not via diffusion through the lipid layer.[21] Or, they may allude to transfection agents, where a small fraction of DNA complexes reach the cytosol, but not by diffusion through the membrane.[8,9] Or they may refer to the (non-controversial) insertion of hydrophobic particles in the lipid membrane.[22] In this way, they would offer a credible foundation for any claims that follow. Just as the unicorn is made more likely because it is a simple and plausible construction of real animals, for example, other land mammals exist that are broadly similar to unicorns (horses), some mammals have horns (rhinos), while some have tusks that twist (narwhals).

Second, authors make a case for the *importance* of the research very often by listing applications of nanomaterials in biology and medicine. This case is often made by citing chemistry articles reporting on the development of nanoparticles and making promises of future applications.[23,24] and not articles reporting applications in biology and medicine. Promises abound and more research is always necessary. It is, therefore, easy to write such introductions. The use of cumulative research, such as in the aggregation of citations to defend a claim about causal relations, plays a crucial role in building scientific theory. It is not without limitations, however (cf. research on meta-analysis in social and psychological sciences[25,26]), and it is important to note that the culmination of *knowledge*, which can advance the field, is not the same as an accumulation of citations that point to beliefs or expectations about highly speculative applications. Especially where these speculative accounts support further speculation. This latter approach can tend towards a "path dependency" process of knowledge building, described by Peacock as a process of knowledge construction dependent on "historical contingencies." [27] The examples we consider in this paper are particularly problematic because the path-dependent approach relies on the accumulation of highly speculative unicorn applications, which are, in turn, used to support further claims for unicorn applications. All of which brings into question the validity (and justifiability) of the scientific knowledge that such approaches generate.[27]

Third, authors may claim that there is *experimental evidence* for the diffusion of nanoparticles through membranes. However, a non-exhaustive analysis reveals two distinct patterns: (1) articles where the claim is presented as common knowledge [7,10–12], such that it is not backed up by any reference to experimental evidence; and (2) articles where the introduction gives the impression that such experimental evidence exists but, on closer inspection, that is not what the references provided show.[8,9,13,22,28,29] [9,22,29] There are also several theoretical articles[30–34] devoted to modeling stripy nanoparticle diffusion through membranes, but both the existence of those structures and their special properties are disputed.[35–37] As Russell notes, "a true belief is not knowledge when it is deduced from a false belief." [38] This is especially important for analyzing common knowledge accounts that benefit from uncertain foundations and weak analytical processes of construction, as we demonstrate here.

Finally, authors will conclude their introduction by justifying their work in relation to other publications studying the nanoparticle-diffuse-through-membrane unicorn by noting that all those previous theoretical efforts have shortcomings, so more research is necessary.

How theoretical models sustain unicorns?

On the one hand, a model serves as useful means to arrive at probabilistic laws, for instance, to represent situations where nature is reliable.[39] A material model is helpful in describing something in physical terms, such as when snooker balls are employed in a demonstration of relations. Meanwhile, a formal model can be used to describe a process that may not have a specific object or property, for instance, by showing a brain structure as modeled on a computer.[40] But the *logical* possibility of any of these models does not guarantee the *physical* possibility of the demonstrated idea, such that the thing that is modeled exists. Because a model may show *some necessary* conditions in the relation between objects (such as the conditions that must be present), this is not the same as showing what would be the *sufficient* conditions (i.e., the full remit of necessary conditions) for those relations or objects to actually exist. In other words, to think of something is no guarantee of its existence, just as to think of the unicorn does not mean that a unicorn must physically exist somewhere in the world. Thus, dozens of theoretical articles about nanoparticles permeating through membranes does not mean that the phenomenon occurs in reality; yet, they assume and reinforce the impression and illusion that it does.

Nevertheless, a model offers the possibility for the scientific and the *verifiable*. It presents ideas that are quantifiable and objective and promises something unambiguous and ascertainable. Yet a model is only as good as the data and the method of its construction. More than this, while a model contains the possibility for testing and experimentation, it remains vulnerable to the accuracy of the content as understood, interpreted, and depicted by researchers. This is similar to a more or less *accurate map* depending on multiple factors, including scaling, interpretation, boundaries (often political), and the selection or omission of data deemed to be more or less useful or valuable, or pertinent to a particular need or situation. As Quine explains, the totality of human knowledge, as well as beliefs, is "a man-made fabric which impinges on experience only along the edges," such that "science is like a field of force the boundary conditions of which are experience." [41] In other words, our experiences, choices, and perceptions all have a role to play in the data that we select and the model that we represent. That is particularly evident in the case of the nanoparticles-diffuse-through-membrane theoretical articles: when the models do refer to physical reality, it is often through the selection of anecdotal results in support of the unicorn while ignoring the overwhelming evidence accumulated over several decades (Figure 2) that nanoparticles enter cells by endocytosis.

The utility of a model always remains to be seen. Kant (A244/B302) describes this when he suggests that we cannot substitute the mere *logical possibility* of an idea or concept, namely that the concept *does not contradict itself*, for the *transcendental possibility* of a thing, namely, that there is or ought to be an object that corresponds to the concept.[42] To confuse these would be to make a category mistake, whereby you confuse the category of "logically possible" with what must be the case. We suggest that this type of error is common. The large number of articles providing models of nanoparticle diffusion through membranes (the logically possible) sustain the mistaken idea that this phenomenon is real.

A scientific theory must make predictions of parameters that can be measured experimentally, thus providing stringent validity tests. The more tests a theory satisfies without being falsified, the more our confidence in it can legitimately increase. However, some scientific articles introduce theoretical models that propose a mechanistic interpretation of a phenomenon without confrontation with experiments. This engenders the kind of conceptual errors that we describe above. That a paper includes experimental results does not guarantee that other pitfalls are avoided. Biases, for example, are unavoidable. Especially as biases include the processes by which researchers select, discard, and evaluate data, as well as how data are reported.[43] Confirmation bias among researchers arises from a very human tendency to prioritize confirmatory details and to have preferences related to the success of one's work.[44] Plus, there is what Cairney describes as the "social dimension to scientific popularity and endurance," which includes the fashionability of concepts.[45]

CONCLUSION

The above analysis illustrates how scientific articles can contribute to sustain deep misconceptions or unicorns. We predict that a systematic analysis (beyond the scope of this perspective) would find that it is very common for introductions to confuse applications of nanomaterials with promises of such applications by citing articles making promises as evidence of the existence of those applications. We suggest that theoretical models of unicorns reinforce belief in their existence (even when the models do not match with experimental evidence), a category error that confuses the logical possibility of something with its existence. In closing, we come back to another source of confusion; ambiguity in terms. What is meant when authors say that nanoparticles diffuse through membranes? The usual meaning of such a sentence is that the objects belonging to the category "nanoparticle" have the property of diffusing through membranes. If, as is usually the case, one means objects which have at least one dimension smaller than 100 nm, then we can say with certainty that nanoparticles do not cross membranes by diffusion through membranes (see, for example, Figure 2). The property of being in the nano-size range does not imply the ability to diffuse through membranes. The nano-size range includes an enormously broad ensemble of objects with extremely varied properties. Thus, for example, the hydrophobic macromolecules described as nanoparticles by Liu et al.,[7] are entirely different from hydrophilic gold nanoparticles studied by Banerji and Hayes.[6] To improve our understanding, it is more helpful to establish how properties such as permeability vary as a function of the specific characteristics (including size) of molecules or particles rather than attempt to define the unique properties of nanoparticles, a category of objects that is so broad that it cannot have any other special property than being nanoparticulate.

Conflict of Interests

The authors declare no conflicts of interest. For a signed statement, please contact the journal office: editor@precisionnanomedicine.com

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REFERENCES

1. Wang, L., Sofer, Z. & Pumera, M. Will Any Crap We Put into Graphene Increase Its Electrocatalytic Effect? ACS Nano (2020) doi:10.1021/acsnano.9b00184.
2. https://commons.wikimedia.org/wiki/File:Tapisserie_d%27Aubusson_les_licornes.jpg. By Anglards-de-Salers - Own work, https://commons.wikimedia.org/w/index.php?curid=49642580 CC BY-SA 4.0
3. Epstein, M. A., Hummeler, K. & Berkaloff, A. The entry and distribution of herpes virus and colloidal gold in HELA cells after contact in suspension. J Exp Med 119, 291 (1964).
4. Harford, C. G., Hamlin, A. & Parker, E. Electron microscopy of HeLa cells after the ingestion of colloidal gold. J Biophys Biochem Cytol 3, 749–756 (1957).
5. Geiser, M. et al. Ultrafine Particles Cross Cellular Membranes by Nonphagocytic Mechanisms in Lungs and in Cultured Cells. Environmental Health Perspectives 113, 1555–1560 (2005).
6. Banerji, S. K. & Hayes, M. A. Examination of Nonendocytotic Bulk Transport of Nanoparticles Across Phospholipid Membranes. Langmuir 23, 3305–3313 (2007).
7. Liu, C. et al. Predicting the Time of Entry of Nanoparticles in Lipid Membranes. ACS Nano 13, 10221–10232 (2019).
8. Lin, J., Zhang, H., Chen, Z. & Zheng, Y. Penetration of Lipid Membranes by Gold Nanoparticles: Insights into Cellular Uptake, Cytotoxicity, and Their Relationship. ACS Nano 4, 5421–5429 (2010).
9. Levy, Raphael. Comment on 'Penetration of lipid membranes by gold nanoparticles: insights into cellular uptake, cytotoxicity, and their relationship'. PubPeer https://pubpeer.com/publications/6593DEE8E72440DEA3AE5AB8BB9374 (2019).
10. Quan, X., Zhao, D., Li, L. & Zhou, J. Understanding the Cellular Uptake of pH-Responsive Zwitterionic Gold Nanoparticles: A Computer Simulation Study. Langmuir 33, 14480–14489 (2017).
11. Lunnoo, T., Assawakhajornsak, J., Ruangchai, S. & Puangmali, T. Role of Surface Functionalization on Cellular Uptake of AuNPs Characterized by Computational Microscopy. J. Phys. Chem. B 124, 1898–1908 (2020).
12. Gupta, R. & Rai, B. Penetration of Gold Nanoparticles through Human Skin: Unraveling Its Mechanisms at the Molecular Scale. J. Phys. Chem. B 120, 7133–7142 (2016).
13. Oroskar, P. A., Jameson, C. J. & Murad, S. Surface-Functionalized Nanoparticle Permeation Triggers Lipid Displacement and Water and Ion Leakage. Langmuir 31, 1074–1085 (2015).
14. Nature editors: all hat and no cattle. PubPeer https://pubpeer.com/topics/1/AE11BE44CF3C40A558F3B453BF53C7 (2016).

15. Lévy, R. PNAS: "your letter does not contribute significantly to the discussion of this paper". Rapha-z-lab https://raphazlab.wordpress.com/2015/11/16/pnas-your-letter-does-not-contribute-significantly-to-the-discussion-of-this-paper/ (2015).

16. Trebino, R. How to Publish a Scientific Comment in 1 2 3 Easy Steps. https://frog.gatech.edu/Pubs/How-to-Publish-a-Scientific-Comment-in-123-Easy-Steps.pdf (2009).

17. Allison, D. B., Brown, A. W., George, B. J. & Kaiser, K. A. Reproducibility: A tragedy of errors. Nature News 530, 27 (2016).

18. Priest, G. What Is Philosophy? Philosophy 81, 189–207 (2006).

19. Dotson, K. Concrete Flowers: Contemplating the Profession of Philosophy. Hypatia 26, 403–409 (2011).

20. Laplane, L. et al. Opinion: Why science needs philosophy. PNAS 116, 3948–3952 (2019).

21. Kell, D. B. & Oliver, S. G. How drugs get into cells: tested and testable predictions to help discriminate between transporter-mediated uptake and lipoidal bilayer diffusion. Frontiers in Pharmacology 5, 231 (2014).

22. Levy, Raphael. Comment on 'Surface-functionalized nanoparticle permeation triggers lipid displacement and water and ion leakage,' PubPeer https://pubpeer.com/publications/4BE3E3C838E137FA51FDC5A5F26017 (2020).

23. Jones, R. The economy of promises. Nature Nanotech 3, 65–66 (2008).

24. Levy, Raphael. Do nanoparticles cross membranes? Further thoughts on 'Time of entry of nanoparticles in lipid membranes.' Rapha-z-lab https://raphazlab.wordpress.com/2020/06/15/further-thoughts-on-the-time-of-entry-of-nanoparticles-in-lipid-membranes-article/ (2020).

25. Hunter, J. E. & Schmidt, F. L. Fixed Effects vs. Random Effects Meta-Analysis Models: Implications for Cumulative Research Knowledge. International Journal of Selection and Assessment 8, 275–292 (2000).

26. Mullen, B., Muellerleile, P. & Bryant, B. Cumulative Meta-Analysis: A Consideration of Indicators of Sufficiency and Stability. Pers Soc Psychol Bull 27, 1450–1462 (2001).

27. Peacock, M. S. Path Dependence in the Production of Scientific Knowledge. Social Epistemology 23, 105–124 (2009).

28. Song, B., Yuan, H., Pham, S. V., Jameson, C. J. & Murad, S. Nanoparticle Permeation Induces Water Penetration, Ion Transport, and Lipid Flip-Flop. Langmuir 28, 16989–17000 (2012).

29. Levy, R. Comment on 'Nanoparticle permeation induces water penetration, ion transport, and lipid flip-flop'. PubPeer https://pubpeer.com/publications/9E1EA2C78086285642179375B4B628 (2020).

30. Li, Y., Li, X., Li, Z. & Gao, H. Surface-structure-regulated penetration of nanoparticles across a cell membrane. Nanoscale 4, 3768–3775 (2012).

31. Van Lehn, R. C. & Alexander-Katz, A. Fusion of Ligand-Coated Nanoparticles with Lipid Bilayers: Effect of Ligand Flexibility. J. Phys. Chem. A 118, 5848–5856 (2014).

32. Van Lehn, R. C. et al. Effect of Particle Diameter and Surface Composition on the Spontaneous Fusion of Monolayer-Protected Gold Nanoparticles with Lipid Bilayers. Nano Lett. 13, 4060–4067 (2013).

33. Gkeka, P., Sarkisov, L. & Angelikopoulos, P. Homogeneous Hydrophobic–Hydrophilic Surface Patterns Enhance Permeation of Nanoparticles through Lipid Membranes. J. Phys. Chem. Lett. 4, 1907–1912 (2013).

34. Levy, Raphael. Are simulations more messy than experiments? (I need help…). Rapha-z-lab https://raphazlab.wordpress.com/2013/08/22/are-simulations-more-messy-than-experiments-i-need-help/ (2013).

35. Cesbron, Y., Shaw, C. P., Birchall, J. P., Free, P. & Lévy, R. Stripy nanoparticles revisited. Small 8, 3714–3719 (2012).

36. Stirling, J. et al. Critical assessment of the evidence for striped nanoparticles. PLoS One 9, e108482 (2014).

37. Verma, A. et al. Surface-structure-regulated cell-membrane penetration by monolayer-protected nanoparticles. Nature Materials 7, 588–595 (2008).

38. Russell, B. The Problems of Philosophy. https://www.gutenberg.org/files/5827/5827-h/5827-h.htm (1912).

39. Cartwright, N. Models: The Blueprints for Laws. Philosophy of Science 64, S292–S303 (1997).

40. Hesse, M. Models and Analogies. in A companion to the philosophy of science (ed. Newton-Smith, W. H.) 299–307 (Blackwell, 2001).

41. Quine, W. V. From a logical point of view: 9 logico-philosophical essays. (Harvard University Press, 1980).

42. Kant, I. Immanuel Kant's Critique of Pure Reason. (Macmillan, 1933).

43. Ioannidis, J. P. A. Contradicted and Initially Stronger Effects in Highly Cited Clinical Research. JAMA 294, 218 (2005).

44. Fanelli, D. "Positive" Results Increase Down the Hierarchy of the Sciences. PLOS ONE 5, e10068 (2010).

45. Cairney, P. Standing on the Shoulders of Giants: How Do We Combine the Insights of Multiple Theories in Public Policy Studies? Policy Studies Journal 41, 1–21 (2013).