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Bernard Delalande [*,†], Hirohisa Tamagawa [†], Vladimir Matveev [†]

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Review

Physical Inconsistencies in the Hodgkin-Huxley Model at the Nanoscale

Bernard Delalande [1,*,†], Hirohisa Tamagawa [2,†] and Vladimir Matveev [3,†]

[1] 280, Avenue de la Pierre Dourdant, 38290 La Verpilliere, France
[2] Department of Mechanical Engineering, Faculty of Engineering, Gifu University, 1-1 Yanagido, Gifu, Gifu, 501-1193 Japan
[3] Laboratory of Cell Physiology, Institute of Cytology, Russian Academy of Sciences, Tikhoretsky Ave 4, St. Petersburg 194064, Russia
* Correspondence: bernard@somasimple.com
† These authors contributed equally to this work.

Abstract

The Hodgkin–Huxley model has provided an extraordinarily successful phenomenological description of the action potential for over seven decades. Its predictive power and mathematical elegance have made it a cornerstone of modern neuroscience. However, the model incorporates mechanistic assumptions about the nanoscale ionic environment of the axonal membrane that were necessarily simplified in 1952 and that modern biophysics allows us to examine critically. In this article, we identify eight independent physical inconsistencies in the mechanistic interpretation of the Hodgkin–Huxley model. These concern: the gel-phase nature of the axoplasm and its consequences for ionic activity; the insufficient ionic reservoir of the peri-membrane volume; the physical implausibility of ionic replenishment at physiological firing rates; ionic congestion and inter-species competition in confined spaces; the reductive representation of ion channels as single conductance parameters; the uncertain relationship between crystallographic channel structures and physiological reality; the osmotic paradox created by intra-pore ionic concentrations; and the systematic physical limitations of patch-clamp recordings. None of these arguments contests the experimental measurements on which the model is based. All of them contest the physical plausibility of the mechanistic interpretation placed on these measurements. The cumulative and mutually reinforcing nature of these inconsistencies suggests that the mechanistic foundations of the Hodgkin–Huxley model deserve serious and systematic re-examination.

Keywords: Hodgkin-Huxley model; action potential; ion channels; peri-membrane volume; axoplasm; nanoscale biophysics; patch-clamp

1. Introduction

The Hodgkin–Huxley model, published in 1952, represents one of the most remarkable achievements in the history of neuroscience [1]. By combining elegant mathematical formalism with meticulous electrophysiological measurements on the squid giant axon, Hodgkin and Huxley produced a quantitative description of the action potential, which remains a cornerstone of modern neuroscience. The predictive power of this model, which earned its authors the Nobel Prize in Physiology or Medicine in 1963, has been validated countless times across diverse excitable cells and species.

The model describes the action potential in terms of voltage-dependent ionic conductances, primarily sodium and potassium, that flow across the cell membrane according to their electrochemical gradients. This framework has proven extraordinarily useful as a phenomenological tool, successfully capturing the essential dynamics of neuronal excitability.

However, the Hodgkin–Huxley model was developed at a time when physical tools to examine the nanoscale environment of the membrane were not yet available. The model necessarily incorporates simplifying assumptions about the ionic environment, the nature of ion channels, and the physical

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properties of the cytoplasm that were reasonable in 1952 but that modern biophysics allows us to examine more critically.

In this article, we do not question the experimental measurements on which the model is based, nor its considerable predictive value at the macroscopic scale. Rather, we examine whether the physical mechanisms assumed by the model are consistent with what is now known about the nanoscale environment of the axonal membrane. We identified eight independent physical inconsistencies that suggest that the mechanistic interpretation of the model deserves careful reconsideration.

2. The Axoplasm Is a Gel: Incorrect Initial Conditions

The Hodgkin–Huxley model treats the intracellular compartment as a homogeneous aqueous solution in which ions are freely diffusible. This assumption, implicit throughout the model, was consistent with the prevailing view of the cytoplasm in 1952. However, substantial evidence accumulated over the following decades demonstrates that this assumption is fundamentally incorrect.

The axoplasm is not a liquid. It is a gel.

This has been extensively documented, notably in the pioneering work of Gilbert Ling on the association-induction hypothesis [2] and more recently by Gerald Pollack, whose work on structured water and gel-phase cytoplasm has provided compelling biophysical evidence for this view [3]. The axoplasm contains a dense network of cytoskeletal proteins—neurofilaments, microtubules, actin—that create a structured matrix throughout the intracellular space.

This has profound consequences for ionic behaviour. In a gel matrix, the ions are not freely diffusible as in bulk solution. A significant fraction of intracellular ions, particularly potassium, are associated with charged protein residues rather than existing as free ions in solution [4]. Therefore, the biochemically measured concentration of K^+ does not reflect the concentration of free mobile K^+ available to participate in electrochemical gradients.

This distinction is critical. The Nernst equation, on which the Goldman–Hodgkin-Katz equation and the Hodgkin–Huxley model are ultimately founded, applies to the *activity* of free ions, not to the total ionic concentration. In a gel phase, the ionic activity can differ substantially from the ionic concentration [5,6].

The assumption of a freely diffusible intracellular ionic pool is therefore not a minor simplification. It is a foundational condition of the model that modern biophysics calls into question before any other consideration is raised.

All subsequent arguments in this article are developed within this framework—and are, if anything, strengthened by it.

3. The Peri-Membrane Volume: An Insufficient Ionic Reservoir

The Hodgkin–Huxley model assumes that the ionic concentrations remain constant on both sides of the membrane throughout the action potential. This is a boundary condition that is never explicitly justified in the original model nor in its subsequent developments.

Modern electron microscopy and structural biology allow us to examine this assumption quantitatively.

3.1. The Available Volume

The functional peri-membrane space, the volume immediately adjacent to the membrane within which ions must be available to enter voltage-gated channels, can be estimated at approximately 12 nm thick, consistent with ultrastructural measurements of the peri-neuronal space reporting values of 10 to 15 nm [7].

For a membrane patch corresponding to the territory of a single ion channel— approximately $100 \times 100\ \text{nm}^2$, consistent with documented channel densities of $\sim$100 channels/μm^2 on unmyelinated axons [8]—the available volume is:

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$$V = 100 \times 100 \times 12\ \text{nm}^3 = 1.2 \times 10^5\ \text{nm}^3 = 1.2 \times 10^{-22}\ \text{L} \tag{1}$$

3.2. The Available Ions

At physiological extracellular Na^+ concentration of 145 mM:

$$N = 145 \times 10^{-3} \times 6.02 \times 10^{23} \times 1.2 \times 10^{-22} \approx 10 \text{ ions per channel territory} \tag{2}$$

3.3. The Required Ions

A single voltage-gated Na^+ channel carries a peak current of approximately 1–4 pA during ∼0.5 ms [8,19]. Taking a conservative value of 2 pA:

$$Q = I \times t = 2 \times 10^{-12} \times 0.5 \times 10^{-3} = 10^{-15}\ \text{C} \tag{3}$$

$$N = \frac{Q}{e} = \frac{10^{-15}}{1.6 \times 10^{-19}} \approx 6000 \text{ ions per action potential} \tag{4}$$

3.4. The Discrepancy

The local ionic reservoir contains approximately 10 ions. Each channel requires approximately 6000 ions per action potential. The discrepancy is three orders of magnitude.

The Hodgkin–Huxley model implicitly assumes that this deficit is continuously compensated for by diffusion from the bulk extracellular medium. This assumption is never stated, never justified, and, as we examine in the following section, is physically problematic.

4. Ionic Replenishment: An Unjustified Assumption

The previous section established that the local ionic reservoir available to a single voltage-gated channel contains approximately 10 ions, while each action potential requires approximately 6000 ions per channel. The Hodgkin–Huxley model implicitly assumes that this deficit is continuously compensated for by diffusion from the bulk extracellular medium. We examine here whether this assumption is physically tenable.

4.1. Diffusion Timescales

The characteristic time for diffusion over a distance x is given by:

$$t \approx \frac{x^2}{2D} \tag{5}$$

For Na^+ ($D \approx 1.33 \times 10^{-9}\ \text{m}^2/\text{s}$) over 12 nm:

$$t \approx \frac{(12 \times 10^{-9})^2}{2 \times 1.33 \times 10^{-9}} \approx 70\ \text{ns} \tag{6}$$

This appears fast relative to the action potential timescale of ∼1 ms. However, this calculation assumes an infinite and immediately available bulk reservoir at constant concentration—precisely the assumption under examination.

4.2. The Origin of the Bulk Reservoir

The nearest source of ionic replenishment beyond the peri-membrane volume is the bulk extracellular medium, itself ultimately supplied by capillary circulation. Between the capillary wall and the axonal membrane, transport is exclusively diffusive—no convection operates at this scale.

For diffusion over a typical capillary-to-axon distance of ∼100 nm:

$$t \approx \frac{(100 \times 10^{-9})^2}{2 \times 1.33 \times 10^{-9}} \approx 4\ \text{ms} \tag{7}$$

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This is four times longer than a single action potential.

4.3. Geometric Tortuosity

The extracellular space surrounding an axon is not a free aqueous medium. It is occupied by extracellular matrix proteins, glial cell processes, adjacent axonal membranes, and membrane-associated proteins, including the channels themselves.

Diffusion in a tortuous medium is reduced by a tortuosity factor τ:

$$D_{\text{eff}} = \frac{D}{\tau^2} \tag{8}$$

In neural tissue, τ is typically 1.5 to 2.5 [9], reducing effective diffusivity by a factor of 2 to 6. The replenishment timescale is therefore 8 to 24 ms per action potential—approaching or exceeding the inter-spike interval at physiological firing rates of 50 Hz.

4.4. The Intracellular Side

The situation is further complicated on the intracellular side by the gel nature of the axoplasm established in Section 2. If intracellular K^+ is substantially associated with cytoskeletal proteins rather than freely diffusible, the effective intracellular reservoir available for rapid replenishment is even more limited than the extracellular one.

The Hodgkin–Huxley model assumes constant ionic concentrations on both sides of the membrane. This assumption, never explicitly stated or justified, is physically untenable at physiological firing rates when the full geometry and physical properties of the peri-membrane environment are considered.

5. Ionic Congestion at the Channel Entrance

The Hodgkin–Huxley model treats each ionic species as independent, moving according to its own electrochemical gradient without interaction with other ionic species or with neighbouring channels. This assumption, inherited from the Goldman-Hodgkin-Katz framework, becomes physically untenable when examined at the nanoscale.

5.1. Electrostatic Attraction of Competing Ions

When a voltage-gated Na^+ channel opens, it creates a local electrostatic potential well that attracts all cations in the immediate vicinity—not exclusively Na^+. In the extracellular peri-membrane volume, the ionic composition is approximately: Na^+ 145 mM, K^+ 5 mM, Ca^{2+} 2 mM, Mg^{2+} 1 mM.

All cationic species respond to the electrostatic field created by the open channel. The channel selectivity filter discriminates between species once they are inside the pore, but does not prevent competing ions from crowding the channel entrance.

5.2. Mutual Repulsion and Inter-Channel Competition

Within the restricted peri-membrane volume, multiple Na^+ ions converging simultaneously toward the same channel entrance will mutually repel each other. This electrostatic repulsion between like-charged ions creates a congestion effect at the pore entrance entirely absent from the Hodgkin–Huxley formulation.

At a channel density of $\sim$100 channels / μ m^2, with multiple channels opening simultaneously during an action potential, the available ionic pool of $\sim$10 ions per channel territory is shared between competing channels, creating a spatial coupling between adjacent channels that the model cannot capture.

5.3. The Dominant Role of Sodium and Ionic Hydration

The overwhelming extracellular dominance of Na^+ (145 mM vs. 5 mM K^+) means that when a K^+ channel opens, Na^+ ions—present at 29 times the concentration of K^+—are preferentially drawn towards the entrance of the pores, competing with and impeding the passage of K^+.

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A further complication arises from ionic hydration shells. In bulk solution, Na^+ carries approximately 4–6 water molecules and K^+ approximately 6–8 water molecules in their primary hydration shell [10]. In the confined peri-membrane space, these hydration shells occupy significant volume and interact with each other, further impeding ionic mobility. The partial dehydration required for the ions to enter the selectivity filter carries an energetic cost of approximately 60–80 kJ/mol for Na^+ [11,12]—never incorporated into the Hodgkin–Huxley conductance formalism.

6. The Ion Channel: A Physically Complex Object Reduced To A Single Parameter

The Hodgkin–Huxley model represents ionic conductances as simple, voltage-dependent parameters. This representation, while mathematically elegant, obscures the extraordinary physical complexity of the objects described.

6.1. Molecular Scale and Residual Polarizations

A voltage-gated Na^+ channel (Nav) comprises approximately 2000 amino acid residues in its primary α-subunit alone, with additional auxiliary β-subunits and extensive glycosylation, for a total atomic count on the order of **50,000 atoms** [13]. This molecular machine is reduced in the Hodgkin–Huxley model to a single conductance parameter and three gating variables.

The charged and polar amino acid residues distributed throughout the channel structure create a complex electrostatic landscape that fluctuates with conformational dynamics, temperature, local ionic environment, and membrane voltage. The Hodgkin–Huxley model captures none of this electrostatic complexity.

6.2. The Crystallographic Problem

Our structural knowledge of ion channels is derived primarily from X-ray crystallography and cryo-electron microscopy [14,15]. These techniques capture a protein that has been extracted from its native membrane, purified, crystallised, or vitrified. The beautiful symmetry observed in these structures reflects a static, averaged, and non-physiological state.

A channel protein embedded in a living membrane at 37°C, surrounded by moving lipids, bathed in ionic solution, subject to a transmembrane electric field and in constant thermal motion, bears an uncertain relationship with its crystallographic image. The mechanistic interpretations built upon these structures are therefore models of a crystallographic artefact as well as of a physiological reality.

6.3. The Water-Ion Sequence

Single-channel recordings and molecular dynamics simulations consistently indicate that ion permeation occurs as an ordered alternation of ions and water molecules in a single file [16,17], a process fundamentally incompatible with the continuous stochastic ionic flux assumed by the Hodgkin–Huxley model.

6.4. The Sodium Block Of Potassium Channels

When a Na^+ ion enters a K^+-selective channel pore—an event whose probability is non-zero given the extracellular dominance of Na^+—it becomes lodged within the selectivity filter, blocking the channel [18]. The mechanism by which such a blocked channel recovers its function is not established. The Hodgkin–Huxley model contains no representation of this blocking phenomenon and its recovery kinetics.

7. The Intra-Pore Concentration: An Osmotic Paradox

Perhaps the most striking physical inconsistency concerns the ionic concentration within the channel pore itself.

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7.1. Calculation

The conducting pore of a voltage-gated Na^+ channel has an approximate diameter of 0.3–0.5 nm at the selectivity filter and a functional length of ∼5 nm, giving a volume of:

$$V = \pi \times (0.25 \times 10^{-9})^2 \times 5 \times 10^{-9} \approx 10^{-28}\ \mathrm{m}^3 = 10^{-25}\ \mathrm{L} \tag{9}$$

With 2–3 ions occupying this volume in single-file permeation:

$$C = \frac{N}{N_A \times V} = \frac{2}{6.02 \times 10^{23} \times 10^{-25}} \approx 33\ \mathrm{mol/L} \tag{10}$$

Table 1. Ionic concentration comparison between extracellular bulk and intra-pore environments.

Environment	Na^+ concentration
Extracellular bulk	0.145 mol/L
Intra-pore	∼33 mol/L
Ratio	∼230×

This concentration of ∼33 mol/L exceeds approximately five times the maximum solubility of NaCl in water (∼6 mol / L at physiological temperature), making it physically impossible to prepare an equivalent bulk solution. The ions within the pore therefore cannot be in a thermodynamic state in which the concepts of ionic concentration, activity, or electrochemical potential — as used in the Nernst and Goldman-Hodgkin-Katz equations — can be meaningfully applied.

7.2. The Electrostatic Containment Paradox

For ions to exist at 33 mol/L within the pore, they must be held there by electrostatic forces of the channel protein sufficiently powerful to maintain this extraordinary concentration against osmotic pressure. However, these same forces must then present an extraordinary barrier to ionic movement through the pore. The Hodgkin–Huxley model never accounts for this osmotic discontinuity of two orders of magnitude. Its conductance formalism implicitly assumes that ions traverse this energetic landscape freely and continuously — an assumption that is physically implausible given the magnitude of the forces involved.

7.3. The Knock-In Knock-Out Mechanism

The standard mechanistic explanation for ion permeation — an incoming ion electrostatically expelling a resident pore ion [8] — requires that the kinetic energy of the incoming ion exceed the binding energy that maintains the resident ion at an effective concentration of 33 mol/L. At physiological temperatures, thermal energy ($kT \approx 25$ meV) may be insufficient to reliably drive this process at the rates required by the model. The cumulative energetic cost of navigating the congested peri-membrane space, competing with other ionic species, shedding the hydration shell, and executing the knock-in knock-out mechanism is never accounted for in the Hodgkin–Huxley framework.

8. The Reliability Of Single-Channel Recordings: Uncontrolled Physical Perturbations

The experimental foundation for the characterisation of ion channels is primarily based on patch-clamp recordings [22], a technique of extraordinary sensitivity. The very sensitivity that makes patch-clamp so powerful also makes it vulnerable to physical perturbations that are rarely acknowledged.

8.1. The Bandwidth Limitation

Patch-clamp amplifiers operate with a finite bandwidth, typically 1–10 kHz in standard configurations. Individual ion transit times through the pores are estimated at 1–10 ns—three to four orders of magnitude faster than the recording bandwidth. The smooth, continuous single-channel currents

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observed in patch-clamp recordings are therefore low-pass-filtered averages of a fundamentally discrete and rapid process. This limitation is not merely theoretical—the patch-clamp literature itself recognises that sub-millisecond gating events are systematically missed, biassing the kinetic constants extracted from such recordings [20,21]. The gating kinetics incorporated into Hodgkin-Huxley-type models are therefore kinetic parameters of a filtered signal, not of the underlying molecular process.

8.2. The Combinatorial Gating Problem

The Hodgkin–Huxley model describes channel gating through independent gating particles whose combined states determine conductance. However, a protein of 50,000 atoms at 37°C exists in a conformational landscape of extraordinary complexity that vastly exceeds the three discrete states assumed by the model. The apparent regularity of gating kinetics may reflect the bandwidth limitation discussed above rather than the intrinsic simplicity of the underlying conformational dynamics.

8.3. Photic Perturbation

Photons in the visible spectrum carry energies of approximately 1.8 to 3.1 eV (170–300 kJ/mol), while conformational energy differences between protein states are typically 1–10 kJ / mol and the thermal energy at 37°C is approximately 2.6 kJ / mol. Visible photons therefore carry energies 17 to 100 times the conformational energy differences between channel states. Membrane proteins contain aromatic residues—tryptophan, tyrosine, phenylalanine—that absorb in the near-UV and visible range. The possibility that ambient laboratory illumination induces conformational perturbations in ion channels during patch-clamp recordings has never been systematically investigated.

8.4. Mechanical and Electromagnetic Perturbation

Patch-clamp recordings operate on the picoampere scale (10^{-12} A). The human body generates mechanical vibrations through cardiac pulsation and respiration, a body surface potential of approximately $\pm$100 mV fluctuating with cardiac and respiratory cycles, and electromagnetic radiation across a broad frequency spectrum. Standard setups employ vibration isolation and Faraday cages, but attenuation is finite. The residual coupling between the observer's physiological signals and the recording system at the picoampere scale cannot be assumed to be zero.

8.5. The Ensemble Averaging Problem

The conductance parameters of the Hodgkin–Huxley model are derived from ensemble averages over many recordings, many channels, many cells, different laboratories, different species, and different experimental conditions. Each layer of averaging suppresses variability and produces smoother parameters. These are then incorporated into the model as fixed constants—as if they represented precise physical properties of a well-defined molecular machine rather than statistical summaries of noisy, filtered, perturbed measurements.

9. Discussion

The arguments developed in the preceding sections share a common structure. In each case, we accept without modification the experimental measurements on which the Hodgkin–Huxley model is founded. Our critique is not of the data, but of the physical assumptions that connect the data to the model.

9.1. The Cumulative Nature of the Inconsistencies

Each argument presented in this article is independent. The gel nature of the axoplasm does not depend on the intra-pore concentration argument. The ionic replenishment problem does not depend on the crystallographic critique. This independence is significant: a model that faces a single anomaly may invoke a specific correction, but a model that faces eight independent physical inconsistencies faces a more fundamental challenge.

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Inconsistencies are, furthermore, cumulative in their effect. The gel axoplasm reduces the effective intracellular ionic reservoir. Insufficient peri-membrane volume limits extracellular availability. The tortuosity of the extracellular space slows the replenishment. Congestion at the entrances of the channels reduces effective flux. The osmotic discontinuity at the entrance of the pores increases the energetic cost of permeation. Each effect compounds the others.

9.2. The Phenomenological Success of HH

It is essential to acknowledge what the Hodgkin–Huxley model achieves. Its mathematical description of the action potential matches the experimental observations with remarkable fidelity. This predictive success is real and should not be minimised.

However, science offers instructive examples of models that achieve excellent phenomenological predictions through mechanisms that have been subsequently shown to be incorrect. The Ptolemaic system predicted planetary positions with considerable accuracy through epicycles that had no physical reality. The predictive success of the Hodgkin–Huxley model establishes that it captures something real about the dynamics of neuronal excitability. It does not establish that its mechanistic assumptions are correct at the nanoscale.

9.3. The Relationship to Alternative Frameworks

The physical inconsistencies identified in this article are not without precedent. Gilbert Ling's association-induction hypothesis [2] proposed that cellular ionic distributions reflected protein-ion associations rather than Nernst equilibria. Gerald Pollack's work [3] has provided modern biophysical support for the view that the cytoplasm cannot be treated as a bulk aqueous solution. The present article does not advocate for any specific alternative. It argues that the physical inconsistencies in the mechanistic interpretation of the Hodgkin–Huxley model are sufficiently serious to warrant a critical re-examination of its foundations.

9.4. The Path Forward

A mechanistically accurate model of the action potential would need to account for the gel-phase nature of the axoplasm, the finite ionic reservoir of the peri-membrane volume, replenishment dynamics under physiological firing conditions, electrostatic interactions between ionic species in confined spaces, the physical complexity of ion channel proteins, the thermodynamic consequences of intra-pore concentration discontinuities, the discrete nature of ion permeation, and the physical perturbations inherent in measurement techniques. A candidate framework addressing these requirements has been proposed in a companion paper [23], which identifies the phase transition of the polyelectrolyte gel as the primary ionic event and the periaxonal hydraulic wave as the conduction mechanism, providing mechanistic accounts of the threshold, the sigmoid stimulus-response curve and the net heat exchange near-zero of the action potential. Its development and experimental testing require close collaboration between electrophysiologists, structural biologists, physical chemists, and computational physicists and are, we submit, a necessary scientific priority.

10. Conclusions

The Hodgkin–Huxley model stands as one of the great achievements of twentieth century science. In honouring this achievement, we owe it the highest form of scientific respect, which is not uncritical acceptance, but rigorous physical examination.

The present article has applied that examination to the mechanistic assumptions underlying the model. We identified eight independent physical inconsistencies.

1. The axoplasm is a gel, invalidating the assumption of freely diffusible intracellular ions on which the Nernst and Goldman-Hodgkin-Katz equations depend.
2. The peri-membrane ionic reservoir contains ∼10 ions per channel territory, while each action potential requires ∼6000—a discrepancy of three orders of magnitude.

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3. Ionic replenishment is impeded by geometric tortuosity and diffusion timescales that approach or exceed inter-spike intervals at physiological firing rates.
4. Ionic congestion, inter-species competition, and hydration shell interactions in the confined peri-membrane space produce a local ionic dynamics entirely absent from the model.
5. The ion channel proteins of ∼50,000 atoms with complex residual polarizations are represented by a single conductance parameter.
6. Crystallographic channel structures represent static, non-physiological states whose relationship to the dynamic, physiologically active channel is uncertain.
7. The intra-pore ionic concentration of ∼33 mol/L exceeds the maximum solubility of NaCl in water by a factor of five and creates an osmotic discontinuity of two orders of magnitude whose thermodynamic consequences are never taken into account.
8. Patch-clamp recordings are subject to bandwidth limitations, photic perturbation, mechanical and electromagnetic coupling, and ensemble averaging that introduce systematic uncertainties not reflected in the model's parameters.

None of these arguments contests the experimental measurements on which the model is based. All of them contest the physical plausibility of the mechanistic interpretation placed on these measurements.

The Hodgkin-Huxley model will continue to serve neuroscience as an indispensable phenomenological tool. What the present analysis calls into question is not the model's descriptive value but its mechanistic truth, its claim to describe not merely what happens during an action potential, but why and how it happens at the physical level.

Science advances not by protecting its monuments, but by subjecting them to the most rigorous scrutiny that contemporary knowledge allows. We hope that this analysis will contribute to a productive re-examination of the mechanistic foundations of the Hodgkin–Huxley model, one that honours the extraordinary achievement of Hodgkin and Huxley by building, carefully and rigorously, upon it.

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