Memory maintenance by PKMζ — an evolutionary perspective

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Abstract

Long-term memory is believed to be maintained by persistent modifications of synaptic transmission within the neural circuits that mediate behavior. Thus, long-term potentiation (LTP) is widely studied as a potential physiological basis for the persistent enhancement of synaptic strength that might sustain memory. Whereas the molecular mechanisms that initially induce LTP have been extensively characterized, the mechanisms that persistently maintain the potentiation have not. Recently, however, a candidate molecular mechanism linking the maintenance of LTP and the storage of long-term memory has been identified. The persistent activity of the autonomously active, atypical protein kinase C (aPKC) isoform, PKMζ, is both necessary and sufficient for maintaining LTP. Furthermore, blocking PKMζ activity by pharmacological or dominant negative inhibitors disrupts previously stored long-term memories in a variety of neural circuits, including spatial and trace memories in the hippocampus, aversive memories in the basolateral amygdala, appetitive memories in the nucleus accumbens, habit memory in the dorsal lateral striatum, and elementary associations, extinction, and skilled sensorimotor memories in the neocortex. During LTP and memory formation, PKMζ is synthesized de novo as a constitutively active kinase. This molecular mechanism for memory storage is evolutionarily conserved. PKMζ formation through new protein synthesis likely originated in early vertebrates ~500 million years ago during the Cambrian period. Other mechanisms for forming persistently active PKM from aPKC are found in invertebrates, and inhibiting this atypical PKM disrupts long-term memory in the invertebrate model systems Drosophila melanogaster and Aplysia californica. Conversely, overexpressing PKMζ enhances memory in flies and rodents. PKMζ persistently enhances synaptic strength by maintaining increased numbers of AMPA receptors at postsynaptic sites, a mechanism that might have evolved from the general function of aPKC in trafficking membrane proteins to the apical compartment of polarized cells. This mechanism of memory may have had adaptive advantages because it is both stable and reversible, as demonstrated by the downregulation of experience-dependent, long-term increases in PKMζ after extinction and reconsolidation blockade that attenuate learned behavior. Thus, PKMζ, the “working end” of LTP, is a component of an evolutionarily conserved molecular mechanism for the persistent, yet flexible storage of long-term memory.

Keywords: PKM zeta, PKMzeta, LTP, Memory
understanding these mechanisms were the molecules that control synaptic plasticity [7].

The form of synaptic plasticity most widely studied in mammalian systems is long-term potentiation (LTP), a persistent synaptic enhancement first characterized in the hippocampus by Bliss and Lømo [8,9]. Interest in LTP grew rapidly with the discovery that the activation of the N-methyl-D-aspartate receptor (NMDAR) triggers both hippocampal LTP induction [10] and hippocampus-dependent spatial learning [11]. Following these seminal findings, over a hundred signaling molecules downstream of the NMDA receptor were characterized [12]. Some, such as Ca²⁺/calmodulin-dependent protein kinase II (CaMKII), initiate a transient early-LTP [13], whereas others, like mitogen-activated protein kinase (MAPK), participate in the regulation of new protein synthesis that is crucial for the transition from early- to more persistent late-LTP [14,15]. Because many of the signaling molecules important for LTP induction were also implicated within a brief time window of an hour after learning during the initial cellular consolidation of long-term memory, the case that an LTP-like mechanism might mediate the cellular basis of memory grew stronger.

But an essential mechanism for both LTP and long-term memory was missing — a mechanism maintaining the changes in synaptic strength and the learned behavior over time. Although scores of the signaling molecules that were activated during LTP were also found to be functionally important for inducing late-LTP, none had been found necessary for maintaining the potentiation once it had been established for 1–2 hours [12,16]. Because inhibitors of protein synthesis applied during this initial time window blocked the induction of both late-LTP and many forms of long-term memory [17,18], the general assumption in the field was that newly synthesized proteins were critical for the persistence of LTP and memory, most likely to serve as building blocks for new synapses. Once these new, experience-dependent synapses had been constructed, however, they could not be eliminated by any enzymatic inhibitor. Thus, in the prevailing theory, long-term memory could be prevented from forming, but could not be erased.

In 2002, however, a brain-specific, autonomously active isozyme of PKC, PKMζ, was found to be both necessary and sufficient for maintaining the late-phase of synaptic potentiation in hippocampal slices [16]. Douglas Ling, Larry Benardo, and our colleagues showed that synapses were potentiated by intracellular perfusion of PKMζ and late-LTP was reversed by inhibiting the kinase, even when the inhibitors were applied many hours after the initial protein synthesis-dependent time window [16,19-22]. Then in 2006, André Fenton and our colleagues showed that the PKMζ inhibitor, zeta inhibitory peptide (ZIP), which effectively blocks the action of PKMζ both biochemically in vitro and within neurons [16,19,23], reverses LTP in vivo 1 day after induction and disrupts spatial memory in the rat hippocampus 1 day or even 1 month after training [22]. The following year, Yadin Dudai and our colleagues began a series of studies showing both ZIP and dominant negative mutations of PKMζ disrupt long-term memory in rat neocortex, up to 3 months after training [24-26].

Subsequently, many forms of long-term memory in a wide variety of neural circuits were shown to be maintained by the persistent activity of PKMζ. In addition to different types of spatial long-term memories [27,28], trace memories in the hippocampus [21], aversive memories in the basolateral amygdala (BLA) [27,29-32], appetitive memories in the nucleus accumbens [33-35], habit memory in the dorsal lateral striatum [36], and elementary associations [24-26,37], extinction [38], and skilled sensorimotor memories [39] in the neocortex were all disrupted by inhibiting PKMζ. Persistent experience-dependent enhancement of synaptic transmission in the hippocampus [21] and the primary visual cortex [40] were also erased by inhibiting PKMζ. Providing an underlying cellular basis for spatial memory erasure, recent work has shown that inhibiting PKMζ disrupts the stable firing patterns of hippocampal place cells exposed to a familiar environment [41]. After the drug has been eliminated, the same place cells establish new stable firing patterns in the familiar environment that have no relationship to the old patterns that had been erased. Some forms of memory were not erased by inhibiting PKMζ including short-term memories mediated by the hippocampus [22] and neocortex [26], and certain long-term memories characterized by the habituation of behavioral responses, such as latent inhibition and attenuation of neophobia [24].

In addition to physiological memory storage, the persistence of several neurological and psychiatric disorders that had been hypothesized to be mediated, in part, by LTP-like changes in the neural circuitry mediating pain or reward was also found to be maintained by PKMζ in animal models. Thus, ZIP ameliorates chronic neuropathic pain when injected in the anterior cingulate cortex [42-44] and spinal cord [45-48], post-traumatic stress disorder in the insular cortex [49], and addiction in nucleus accumbens [33-35], BLA [38], hippocampus [50], and ventral tegmental nucleus [51]. Abnormal aggregations of PKMζ are also observed in and near neurofibrillary tangles in the brains of individuals with Alzheimer’s disease [52].

ZIP, a cell-permeable pseudosubstrate peptide inhibitor, is the most commonly used pharmacological tool to inhibit PKMζ. ZIP applied extracellularly to neurons
blocks the action of PKMζ perfused into CA1 pyramidal cells in hippocampal slices [19,23], PKMζ transfected into primary cultured hippocampal neurons [53], and PKCaζ introduced into sensory neurons [47]. The IC50 of the ability of ZIP to inhibit PKMζ-mediated potentiation of α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid receptor (AMPAR) responses at synapses of CA1 pyramidal cells is nearly identical to the IC50 of its ability to reverse late-LTP at these synapses [19]. Because both full-length atypical PKC (aPKC) isoforms, PKCaζ and PKCaζ/λ, contain the identical pseudosubstrate sequence, ZIP is also a standard reagent to inhibit the function of full-length aPKC within cells [54] and to identify intracellular aPKC substrates [55]. One paper had suggested ZIP at the doses used to inhibit PKMζ postsynaptically perfused into neurons was not effective on a PKMζ fusion protein overexpressed in cultured cells [56]. These negative results, however, were subsequently explained to be a consequence of using the standard doses of ZIP in overexpression systems that increase kinase levels between 1–2 orders of magnitude above normal [23]. At such high levels of overexpression, the exogenous “spare” kinase, analogous to spare receptors, far exceeds the endogenous kinase, and the standard doses of ZIP that inhibit PKMζ in neurons and reverse LTP maintenance would be expected to have no noticeable effect [23].

Extending beyond maintenance to expression, Karim Nader and our colleagues at McGill University showed that PKMζ sustained late-LTP and long-term memory by a common mechanism of synaptic enhancement. PKMζ potentiates synaptic transmission by modifying the trafficking of GluA2 subunit-containing AMPARs so as to increase the number of receptors at postsynaptic sites [30,57,58] (Figure 1). Nader and our colleagues showed that blockers of GluA2 endocytosis prevent the disruption of previously stored memory by the standard dose of ZIP lower than that employed in previous studies on the disruption of fear conditioning that had tested memory retention a day after injection of the drug [30]. In a subsequent study of the retention of a learned active avoidance response using the standard dose of ZIP in the BLA, however, confirmed persistent amnesia for a week after drug injection [31], demonstrating that memory erase by ZIP in the BLA was consistent with that specifically erases the long-term memory trace maintained by these structures.

Because the half-life of ZIP is ~2 hours [32], initial studies on the disruption of fear conditioning that had tested memory retention a day after injection of the drug into the BLA had indicated that the persistence of memory erasure in the BLA would be similar to that in hippocampus and neocortex [30]. One paper, using a dose of ZIP lower than that employed in previous studies of other brain regions, suggested that the memory loss for fear-potentiated startle was temporary [60], although see the discussions in references [61,62]. A subsequent study of the retention of a learned active avoidance response using the standard dose of ZIP in the BLA, however, confirmed persistent amnesia for a week after drug injection [31], demonstrating that memory erase by ZIP in the BLA was consistent with that
observed in other brain regions. Interestingly, in the study that had used the low dose of ZIP, memory loss persisted when the rats were reexposed to the CS alone, a day after drug infusion [60]. Because the standard dose of ZIP erases multiple memories within a brain region, with or without CS reactivation [24], if low dose ZIP selectively disrupts the CS-US association of the specific reactivated CS, such doses of the drug might be used to erase specific memories, similar to the specific disruption of reactivated memory that is the hallmark of reconsolidation blockade [63].

The molecular mechanisms of synaptic memory storage by PKMζ

PKMζ’s ability to store experience-dependent information at synapses is due to its unique structure as an autonomously active form of PKC [64-66]. Full-length PKC isoforms are activated by conformational changes induced by second messengers [67]. Each PKC consists of an N-terminal regulatory domain linked by a hinge region to a C-terminal catalytic domain. Under basal conditions in the cell cytosol, full-length PKCs are inactive because an autoinhibitory pseudosubstrate within the regulatory domain interacts with and blocks the catalytic domain. Second messengers stimulate the full-length PKCs by binding to the regulatory domain, translocating the enzyme to membrane, and inducing a conformational change that releases the autoinhibition. This allosteric mechanism activates all three classes of PKC isoforms — conventional PKCs by Ca2+; novel PKCs by DAG, but not Ca2+; and atypical PKCs, including the full-length PKCζ, by neither Ca2+ nor DAG, but by alternate lipid second messengers and proteins that bind to the aPKC regulatory domain. Because the second messengers that activate the full-length PKCs are generally short-lived, this mechanism of action is transient and rapidly reversible, allowing PKC to participate in multiple rounds of short-term signal transduction.

In contrast to the full-length isoforms, PKMζ consists of a PKC catalytic domain without a regulatory domain [64,65]. Lacking the autoinhibitory pseudosubstrate of the PKCζ regulatory domain (i.e., the amino acid sequence of ZIP), PKMζ is autonomously and thus persistently active [64,66]. The formation of PKM was originally described in biochemical in vitro studies to be through limited proteolysis of full-length PKC at its hinge region, separating the regulatory from the catalytic domain [68]. However, the generation of PKMζ in neurons is by a transcriptional and translational mechanism unique to the ζ gene, PRKCZ, which produces the ζ catalytic domain directly through new protein synthesis [65] (Figure 1). The PRKCZ gene contains two promoters, one upstream of the exons of the N-terminal regulatory domain that generates the full-length PKCζ mRNA and protein, and a second internal promoter within a large intron that produces a PKMζ mRNA, the translation of which begins at an evolutionarily conserved methionine in the hinge region to produce an independent ζ catalytic domain [65]. In the forebrain, PKMζ mRNA is expressed by the PRKCZ gene, and PKCζ mRNA is transcribed only in trace amounts, except in the lateral olfactory tract; in the hindbrain, both mRNA species are transcribed [65,69]. Outside the nervous system, PKCζ mRNA is expressed in various cell-types, and PKMζ mRNA is transcribed only in trace amounts [65].

Under basal conditions in neurons, PKMζ mRNA is transported to dendrites [70] and is translationally repressed by its long 5′untranslated region [65]. Following NMDAR activation that triggers LTP, many of the signaling molecules important for LTP induction, including CaMKII, PKA, MAPK, phosphatidylinositol 3-kinase (PI3K), mammalian target of rapamycin (mTOR), as well as actin filament formation, act in concert to increase PKMζ synthesis [65,71,72] (Figure 1). Brain-derived neurotrophic factor (BDNF) injected into the hippocampus in vivo also increases PKMζ [73] and in theta burst-triggered LTP plays an additional role in decreasing the initial degradation of PKMζ thus further contributing to increases in the kinase [74]. Immediately after translation, the nascent PKMζ is rapidly phosphorylated by phosphoinositide-dependent kinase 1 (PDK1), which locks the PKMζ in a maximally activated state [23,71]. Subsequently, persistent PDK1 phosphorylation is no longer required for the autonomous activity of PKMζ [23]. Whereas increased translation of pre-existing PKMζ mRNA is the mechanism for the formation of PKMζ in LTP [65,71], persistent increases in ζ mRNA also occur after some forms of learning [75], suggesting additional transcriptional regulation of the abundance of the PKMζ message and thus the translational capacity of the neuron to synthesize PKMζ. After synthesis, PKMζ acts as an LTP-specific plasticity-related protein (PRP) that is captured at recently activated synapses that have undergone “synaptic tagging” [17,20,76-79]. CaMKII has been proposed to be a component of the synaptic tag sequestering PKMζ [79].

Once at the synapse, the functional target of PKMζ for synaptic potentiation is the GluA2 subunit of the AMPAR [30,58]. The interaction between PKMζ and GluA2, originally described in rodents, is likely to be evolutionarily conserved, because the colocalization of the two molecules at synaptic sites has recently been observed to positively correlate with memory performance in young and aged non-human primates [80]. Interactions between the trafficking protein N-ethylmaleimide-sensitive factor (NSF) and GluA2, which was originally described as part of a homeostatic mechanism maintaining AMPARs at
synapses [81-84], are critical for the synaptic potentiation by PKMζ, but the precise targets of phosphorylation that mediate the enhancement have not yet been established [30,58]. PKMζ also interacts with the postsynaptic scaffolding protein, kidney and brain expressed protein (KIBRA) [85,86], which has been associated by genetic studies with human memory performance [87], and the C-terminal of PKMζ is a PSD-95/DLG/ZO-1 (PDZ)-binding sequence that interacts with protein interacting with PKC 1 (PICK1) [58]. Both KIBRA and PICK1 also bind to the AMPAR GluA2 subunit and participate in the regulation of the trafficking of the receptor to postsynaptic sites [88-90].

Perhaps related to its role in AMPAR trafficking, PKMζ also increases the aggregation of postsynaptic density protein 95 (PSD-95) at synapses [53], which may be through phosphorylation of the palmitoylation enzyme ZDHHC8 [91]. PKMζ alters the morphology of spines in cultured neurons [92], and the amount of PKMζ in spines positively correlates with the area of the PSD in synapses of the dentate gyrus in non-human primates [80]. Because ZIP reverses the PKMζ-mediated aggregation of PSD-95 within hours of drug application, these structural changes of synapses may, like synaptic potentiation, be maintained by the persistent enzymatic action of PKMζ [53].

Remarkably, a persistently active PKM form is also generated from the invertebrate aPKC, which lacks the vertebrate PKMζ translational start site [93], and this atypical PKM plays fundamental roles in long-term memory maintenance in widely divergent invertebrate phyla. Working with the arthropod Drosophila melanogaster, Jerry Yin and our colleagues at the University of Wisconsin at Madison showed that the persistent activity of atypical PKM is both necessary and sufficient for long-term memory of olfactory avoidance behavior that is induced by associative conditioning [96]. Drosophila atypical PKM is enriched in the fly head [96], just as PKMζ is specifically expressed in neural tissue [65,97], but the mechanism for the formation of atypical PKM in Drosophila has not yet been elucidated. In the mollusk Aplysia californica, David Glanzman and colleagues at UCLA found that the persistent activity of atypical PKM is crucial for maintaining behavioral long-term sensitization of withdrawal reflexes as late as 7 days after training, well beyond the initial, protein synthesis-dependent consolidation phase for the sensitization [98].

A comparative genomic analysis of atypical PKC performed by Wayne Sossin and colleagues at McGill University found that the translational mechanism for the formation of PKMζ, the hallmark of which is a conserved methionine in the hinge region that initiates the synthesis of PKMζ [65], arose around the time of the gene duplication of the single invertebrate aPKC gene into the two vertebrate aPKC isoforms, ζ and η/λ [93]. These two isoforms, whose actions can be similar in neurons [47], are the two most closely related genes of the 9-member PKC gene family. Extending this analysis, Ling Pan (SUNY Downstate) and I found that the lamprey, an early, jawless cyclostome vertebrate, has an apparent single aPKC, with features of both PKCζ and PKCη/λ, that contains the hallmark hinge methionine found in PKMζ that initiates translation of the independent catalytic domain. Therefore, the formation of atypical PKM by new protein synthesis originated at or before the splitting of cyclostomes from the main vertebrate line of evolution (the cyclostome–gnathostome split). This establishes the origin of the formation of PKM by new protein synthesis, and therefore the mechanism maintaining late-LTP, at least ~500 million years ago in the Cambrian period [94,95]. The evolutionary history of PKMζ, LTP, and long-term memory

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transfected PKMζ into the neocortex of rats, Yadin Dudai and our colleagues at the Weizmann Institute showed that not only are new memories strengthened when PKMζ is overexpressed before training, but even old, faded memories are robustly enhanced when the kinase is overexpressed a week after training [25]. The mechanisms by which increasing PKMζ by overexpression enhances memory in both vertebrates and invertebrates are not known, but may involve upregulation of the positive feedback loops of local translation and “synaptic autotagging” that have been proposed to maintain the synaptic compartmentalization of PKMζ [59], as discussed in the next section.

Why is the persistently active PKM form of an atypical PKC crucial for memory maintenance, whether it is generated by cleavage of full-length PKC as in Aplysia, or by transcription from an internal promoter within the PKCζ gene as in vertebrates? Although one can only speculate, a clue may be the original function of aPKC in cells. Single cell organisms such as yeast express a single PKC, but multicellular animals express multiple PKC isoforms generated by gene duplication. In C. elegans, the function of aPKC has already specialized to establish and maintain apical compartments within polarized cells through participation in a highly conserved multiprotein complex, called the anterior PAR complex (for partitioning), consisting of the adapter proteins PAR6 and PAR3, the small GTPase Cdc42, and aPKC [103] (Figure 2A). In this apically localized complex, Cdc42 receives extracellular signals and stimulates PAR6, which then binds to the regulatory domain of aPKC, activating the kinase [104]. The PAR complex is conserved in polarized cells throughout evolution and defines the anterior pole of the C. elegans embryo, the apical domain of Drosophila neuroblasts to control their asymmetric division, and the apical membrane of epithelial cells to promote apical-basal polarity and the formation and maintenance of cell–cell junctions [103,105-107]. Although the mechanisms by which the PAR complex mediate polarity are only beginning to be elucidated, a genome-wide screen in C. elegans has shown that the complex directs the trafficking of membrane proteins through the regulation of endocytosis and vesicle recycling [108,109]. This mechanism is evolutionarily conserved because it is also observed in human HeLa cells [108].

The general function of aPKC to distribute membrane proteins to apical compartments may have adapted to control the trafficking of glutamate receptors to the postsynaptic density, the apical compartment of the synaptic spine (Figure 2B). Atypical PKC may originally have participated in development of the synapse. Indeed, roles for PKMζ in synaptic maturation and dendritic development have recently been described [91,110].

Once established as a mechanism for trafficking glutamate receptors to the synapse during development, the further activation of full-length aPKC might have been useful for short-term synaptic plasticity and short-term memory. Then, mutations that either allow proteolysis in the hinge between the regulatory and catalytic domains in invertebrates [99], or that generate independent translation of the catalytic domain in vertebrates [65], would have transformed this short-term memory mechanism into a long-term memory mechanism (Figure 2B).

The truncation of the aPKC regulatory domain to form an independent catalytic domain would serve two purposes in a molecular mechanism of long-term memory
Because memories up to 3 months old can be erased by PKMζ inhibitors [26] and the amnestic effect of PKMζ inhibition is blocked by GluA2 endocytosis inhibitors [30,34,36], a recent review has proposed that PKMζ and its downstream targets form a self-perpetuating, positive-feedback network through a process of “synaptic auto-tagging” that can persist for months to maintain very long-term memories [59] (Figure 2B). After its local synthesis during LTP or memory formation, PKMζ, by a process involving inhibition of endocytosis through the action of NSF, traffics GluA2-containing AMPARs to the synapse, where AMPAR-binding proteins, such as PICK1 and KIBRA, also bind to PKMζ and thus maintain the kinase at the appropriate postsynaptic sites. Applications of PKMζ inhibitors disrupt the positive-feedback signaling network and erase the potentiated state of the synapse and long-term memory by releasing the GluA2 endocytosis that has been inhibited by the activity of PKMζ. Thus, PKMζ inhibition allows the AMPARs and PKMζ to be eliminated from the appropriate postsynaptic sites. The collapse of the positive feedback network would be rapid, because both LTP [2] and long-term memory [22,27] are disrupted within 2 hours of exposure to ZIP or other PKMζ inhibitors. After the inhibitors have been eliminated, because the original postsynaptic sites of PKMζ formation and subsequent positive feedback have been lost, the potentiation [21] and long-term memory [22,24] are permanently eliminated, and the strength of the synapse and the animal’s behavior reset to the naïve state [59].

Other positive feedback loops involving enhanced local translation have been proposed to maintain increased levels of the kinase at potentiated synapses (Figure 2B). In LTP, for example, PKMζ phosphorylation is required for the increased synthesis of PKMζ [71]. In particular, PKMζ phosphorylation of peptidyl-prolyl cis-trans isomerase NIMA-interacting 1 (PIN1) upregulates local dendritic synthesis and the translation of PKMζ [111] (Figures 1 and 2B). Interestingly, phosphorylation by the Aplysia atypical PKM is required for the formation of the kinase by proteolysis, also forming a positive feedback loop [99]. Thus, different positive feedback loops may have evolved to maintain persistent increased atypical PKM in vertebrates and invertebrates, each specific to the mechanism by which the atypical PKM is generated during the formation of long-term memory.

Although the long-term storage of information by PKMζ is isolated from short-term signaling at the synapse, as discussed in the previous section, the rapid erasure of memory by PKMζ inhibitors suggests the possibility that the long-term information stored by PKMζ might also be modifiable by new experiences. Thus, an animal might quickly update a long-term memory by rapidly degrading the PKMζ molecules restricted to potentiated synapses and disrupting the positive feedback loops maintaining the maladaptive information. The physiological erasure of a long-term memory within a neural circuit by downregulating PKMζ would return the circuit to its naïve state, while preserving the circuitry that had been established during development to mediate specific behaviors. This is because whereas pharmacological or dominant negative inhibitors of PKMζ disrupt LTP maintenance, these inhibitors do not affect basal synaptic transmission either in brain slices or in vivo in mature animals [16,19,22,23,57].

The downregulation of persistent increases of PKMζ has recently been observed during memory extinction and reconsolidation blockade [112]. In animal models of drug addiction, persistent increases of PKMζ maintain drug-craving memory in the neural circuit mediating reward and emotion, including the nucleus accumbens and the BLA [34,38,112]. When memories associated with drug use are partially extinguished by repetitive exposure to the CS, the previously induced, persistent increases of PKMζ in the BLA are reversed, and PKMζ increases in the infralimbic cortex, where the kinase is critical for maintaining extinction [38]. Furthermore, a form of reconsolidation blockade, in which reactivation of the memory is followed by extinction, produces more robust reversal of PKMζ in the BLA, greater increases in the infralimbic cortex, and stronger disruption of the drug-associated memory than extinction alone [112]. Decreases in PKMζ have also been observed in the hippocampus with spatial familiarity [113], which, like extinction, is produced by multiple exposures to an environment without experimental reinforcement. Although the
mechanism for downregulating PKMζ during the repetitive exposure to a stimulus is not known, proteolytic degradation of PKMζ has been observed in the maintenance of NMDAR-dependent long-term depression (LTD) [114,115], suggesting the possibility that LTD or depotentiation induces a persistent loss of PKMζ that maintains familiarity and, in some circuits, the extinction of memory.

Fundamental information for understanding these mechanisms of memory stability and erasure are the half-life of PKMζ and the mechanisms maintaining the compartmentalization of the kinase at specific synapses, such as those proposed in the model of PKMζ synaptic auto-tagging [59]. Although another mechanism of memory storage may take over from PKMζ after 3 months [26], the more parsimonious hypothesis is that PKMζ maintains information for a memory’s lifetime. For humans, how can the fidelity of PKMζ-mediated positive feedback loops be maintained for decades? What is the relationship between the persistence of PKMζ that functionally maintains long-term memory and the structural changes associated with long-term memory, particularly the growth of new synapses [7]? These are essential questions for the future study of PKMζ and memory storage.

Conclusion
The persistent increased activity of PKMζ maintains LTP and perpetuates many, but not all, forms of long-term memory. PKMζ and its invertebrate orthologues provide insight into the evolutionary history of LTP-like synaptic plasticity and long-term memory. The role of atypical PKM in maintaining long-term memory may have emerged as a specialized mechanism for persistently increasing postsynaptic AMPARs from the more general function of aPKC in membrane protein trafficking to the apical compartment of polarized cells. The origin of the mechanism of late-LTP by new synthesis of atypical PKM can be traced to early vertebrates in the Cambrian period. This synaptic information storage mechanism proved capable of recording experiences within neural circuits in a way that was both stable for months, yet reversible as new contingencies arise, and appears to have been useful for animal survival for hundreds of millions of years.

Competing interests
The author has no competing financial interests.

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