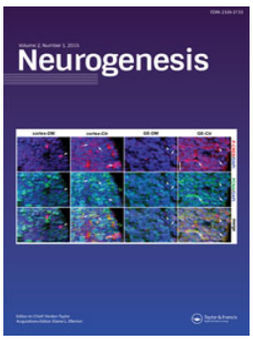

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RESEARCH PAPER

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Rheb1 mediates DISC1-dependent regulation of new neuron development in the adult hippocampus

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Keywords: adult neurogenesis, neurodevelopment, DISC1, Rheb1

A large number of susceptibility genes have been implicated in psychiatric disorders with a developmental origin, yet their biological roles and signaling mechanisms in neurodevelopment are largely unknown. Disrupted-In-Schizophrenia 1 (*DISC1*), a susceptibility gene for several major psychiatric disorders, regulates the development of newborn neurons in the adult hippocampus. Systemic pharmacological inhibition of mTOR signaling with rapamycin has been shown to rescue DISC1 deficiency-induced neurodevelopmental defects, as well as cognitive and affective deficits. Whether mTOR signaling plays a cell-autonomous and/or non-cell-autonomous role in DISC1-dependent regulation of neuronal development is not clear. Here we provide genetic evidence that hyper-activation of mTOR activator Rheb1 (Ras homolog enriched in brain 1) in newborn neurons recapitulates DISC1 deficiency-induced neurodevelopmental defects, including neuronal morphogenesis and migration. We further show that genetic deletion of *Rheb1* rescues those defects in a cell-autonomous fashion in developing newborn neurons in the adult hippocampus. Our genetic and functional studies demonstrate that Rheb1 acts as a key mediator of DISC1-dependent regulation of mTOR signaling and neuronal development during adult hippocampal neurogenesis.

Introduction

One of the greatest challenges in treating complex neurodevelopmental disorders, such as schizophrenia (SCZ) and autism spectrum disorders (ASD), is identifying determinative pathophysiology and potential targets for effective intervention. While genome-wide association studies have revealed a vast number of candidate risk genes associated with SCZ and ASD, much less is known about how these genetic risk factors could impact the developing nervous system.[1-3] Determining the functional role of risk genes in normal neuronal development and identifying their signaling pathways will not only lead to a better understanding of the pathogenesis of SCZ and ASD, but also the identification of potential targets for therapeutic intervention.

Disrupted-In-Schizophrenia 1 (*DISC1*) is a rare risk gene originally identified at the breakpoint of a balanced chromosomal translocation t(1;11)(q42;q14) that co-segregates with major mental illness in a unique Scottish pedigree.[4] Subsequent genetic linkage and association studies identified additional variants of *DISC1* that are associated with SCZ, affective disorders, and ASD.[5,6] *DISC1* encodes a scaffold protein with multiple binding partners that have a broad impact on intracellular signaling pathways and neurodevelopmental processes,[7,8] including neural progenitor proliferation,[9] neuronal migration,[10] axonal and dendritic growth, and synapse formation.[11-16] During adult hippocampal neurogenesis, DISC1 knockdown decreases proliferation in neural progenitors,[9] but accelerates dendritic growth and leads to soma hypertrophy, ectopic dendrites and aberrant

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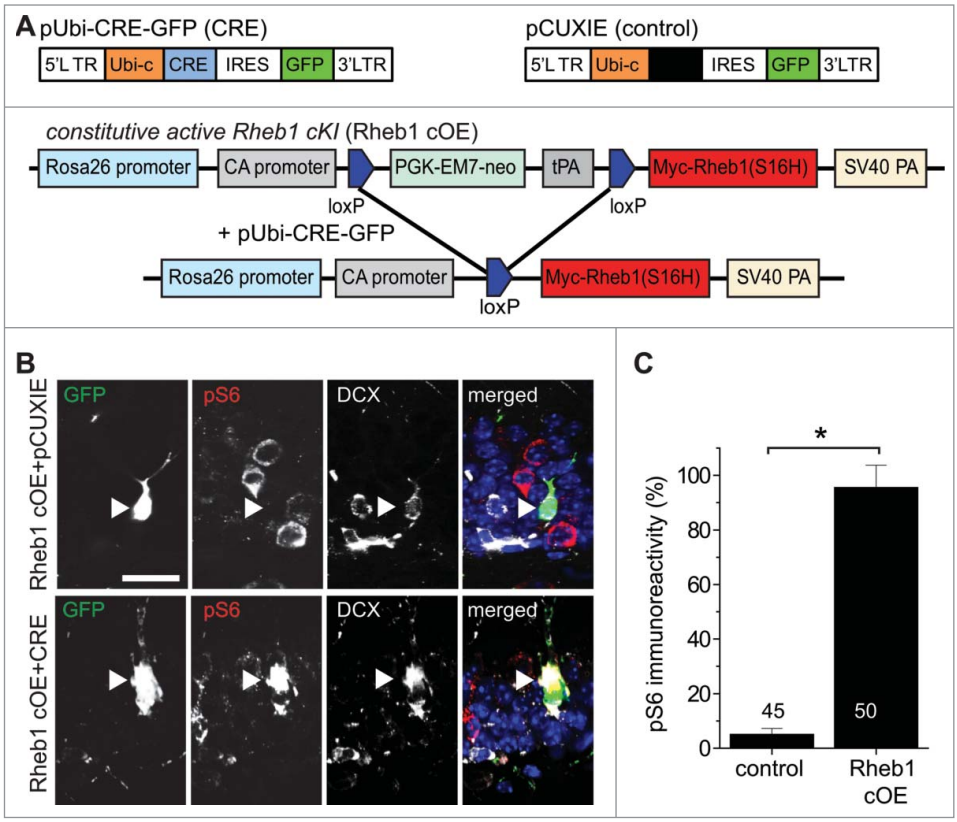

Figure 1. Hyper-activation of Rheb1 is sufficient to activate mTOR signaling in newborn neurons in the adult hippocampus. (**A**) Schematic diagrams of retroviral vectors used for expressing GFP (pCUXIE), co-expressing GFP and Cre recombinase (pUbi-CRE-GFP) (upper panel). Also shown is a schematic diagram for strategies of expressing Rheb1-S16H specifically in proliferating neural progenitors in the adult dentate gyrus (lower panel). Retroviruses co-expressing GFP and Cre were stereotaxically injected into the dentate gyrus of adult *Rheb1* cOE mice. (**B and C**) Increased mTOR signaling in newborn neurons expressing Rheb1-S16H at 14 dpi. Shown in (**B**) are sample confocal images of immunostaining of GFP, pS6-Ser235/236 and immature neuron marker DCX. Arrowheads point to GFP^+DCX^+ newborn neurons. Note the high pS6 levels in a GFP^+ neuron expressing Cre (lower panel), and very low levels in GFP^+ only neurons (upper panel). Scale bar: 20 μm. Shown in (**C**) is a summary of quantification of pS6 levels in GFP^+ newborn neurons under different conditions. The immunofluorescence intensity of pS6 in GFP^+DCX^+ cells was normalized to that of GFP^-DCX^+ neurons in the same brain sample. Numbers associated with the bar graph indicate numbers of GFP^+ neurons examined. Values represent mean + SEM (*: $p < 0.01$; Student's t-test).

positioning of newborn neurons.[11] Interestingly, DISC1 deficiency in hippocampal adult-born neurons alone is sufficient to induce specific behavioral deficits.[17] Converging evidence from biochemical and genetic studies suggest a model in which DISC1 regulates adult-born neuron development through regulation of intracellular Akt activation.[18,19] Notably, systemic inhibition of mTOR signaling by rapamycin during early stages of neuronal development rescues specific behavioral impairments as well as cellular deficits induced by DISC1-deficiency, but rescues only behavior deficits, without changing the cellular phenotype, if applied during later stages.[17] Thus, mTOR signaling may have both a direct and an indirect impact on neuronal development and circuitry function. It is unknown how mTOR is activated in this model and whether mTOR is required in a cell-autonomous manner to mediate DISC1-dependent regulation of neuronal development.

The small GTPase Rheb1 (Ras homolog enriched in the brain 1) has been shown to be a specific activator of mTOR complex 1 via direct binding to mTOR and it stimulates the kinase activity of mTOR when loaded with GTP *in vitro*.[20-23] The role of Rheb1 in the brain has just begun to be elucidated. One recent study showed that Rheb1 is both necessary and sufficient for mTOR complex 1 activation *in vivo* in the brain and that Rheb1 is essential for embryonic brain development and myelination[24] and for hippocampal dependent plasticity.[25] In another study, deletion of Rheb1 in hypothalamic neurons leads to reduced food intake and hypoglycemia, a phenotype also found when hypothalamus mTOR complex 1 activity is dysregulated.[26,27]

Here, using *Rheb1* conditional knockout and conditional overexpression mouse models in combination with retrovirus-mediated expression of Cre to specifically manipulate Rheb1 expression in newborn neurons in the adult hippocampus, we provide evidence that Rheb1 is essential for mTOR signaling and development of newborn neurons. Furthermore, Rheb1 overexpression mimics the cellular phenotypes induced by DISC1 knockdown, while Rheb1 knockout rescues those deficits. Therefore, our studies suggest that Rheb1 is an important regulator of mTOR signaling and a downstream mediator of DISC1 to regulate neuronal development during adult neurogenesis.

Materials and Methods

In vivo birth-dating, genetic manipulation of neural progenitors with retroviruses

Engineered self-inactivating murine onco-retroviruses were used to co-express shRNA under the U6 promoter and a fluorescent marker (GFP or DsRed) under EF1a promoter, or Cre recombinase and GFP under the ubiquitin promoter specifically in proliferating cells and their progeny, as previously described[28] (**Fig. 1A and 2A**). Specific shRNAs against mouse *Disc1* (sh-D1)

and control shRNA have been previously characterized and validated via rescue experiments with shRNA-resistant wild-type DISC1 expression.[11,18,19] High titers of engineered retroviruses were produced by co-transfection of viral vectors and vesicular stomatitis viral envelope (VSVP) into 293gp cells followed by ultra-centrifugation of viral supernatant as previously described.[11]

The following adult mice were used (6–8 weeks old): WT (C57BL/6) mice, *ROSA-Rheb1-S16H*$^{f/f}$ and *Rheb1*$^{f/f}$ mice.[24] Animals housed under standard conditions were anaesthetized and concentrated retroviruses were stereotaxically injected into the dentate gyrus at 4 sites (0.5 μl per site at 0.25 μl/min) with following coordinates (from Bregma in mm): posterior = 2, lateral = + 1.6, ventral = 2.5; posterior = 3, lateral = + 2.6, ventral = 3.2, as previously described.[11] All animal procedures used in this study were performed in accordance with the protocol approved by the Institutional Animal Care and Use Committee.

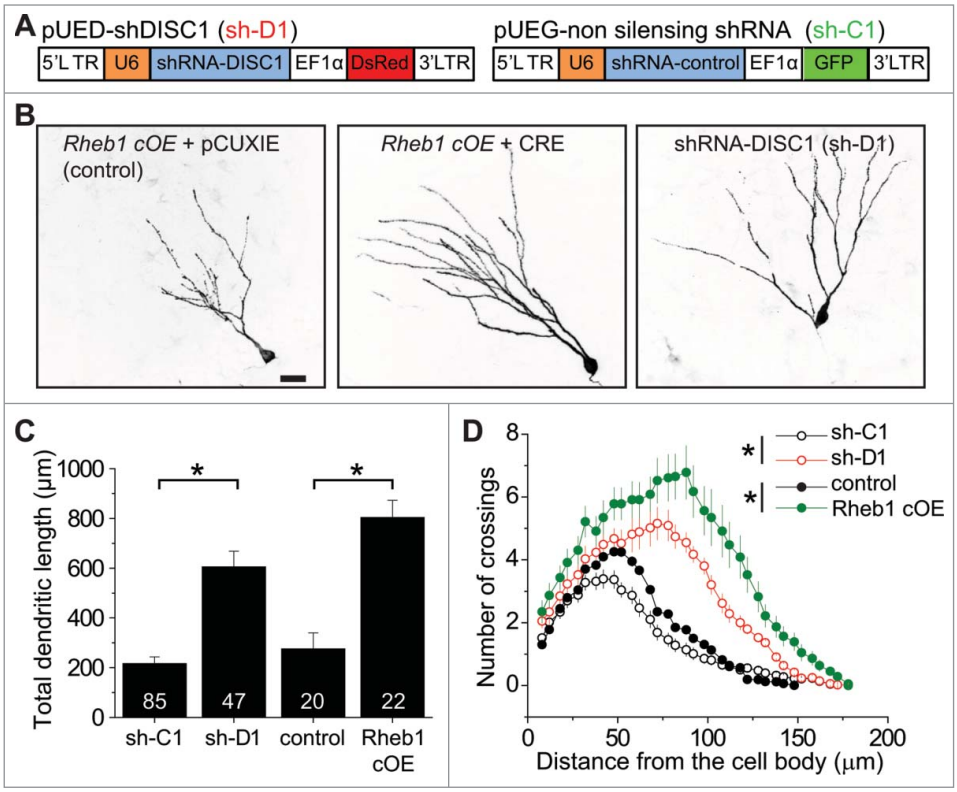

Figure 2. Rheb1 hyper-activation recapitulates DISC1 knockdown-induced dendritic developmental defects of newborn neurons in the adult hippocampus. (**A**) Schematic diagrams of retroviral vectors used for co-expressing GFP and sh-C1 or DsRed and sh-D1. (**B–D**) Dendritic length and complexity of newborn neurons at 14 d post retroviral injection (dpi) to co-express GFP and sh-C1 or sh-D1 in proliferating neural progenitors in the dentate gyrus of adult WT mice, or to express GFP alone (control) or co-express GFP and Cre (*Rheb1* cOE) in conditional Rheb1-S16H overexpression mice. Shown in (**B**) are sample projection confocal images under different conditions. Scale bar: 20 μm. Shown in (**C**) is a summary of total dendritic length. Numbers associated with bar graphs indicate total numbers of GFP$^+$ cells examined from 4 animals for each condition. Values represent mean + SEM. (*: $p < 0.01$; Student's t-test). Shown in (**D**) is a summary of Sholl analysis of dendritic complexity of newborn neurons. The same groups of neurons as in **Figure 2C** were examined. Values represent mean + SEM (*: $p < 0.01$; 2-way ANOVA).

Immunohistology, confocal imaging and analysis

Coronal brain sections (40 μm in thickness) were prepared from injected mice and processed for immunostaining as described.[29,30] The sections were incubated for 30 min in 4′,6′-diaminodino-2-phenylindole (DAPI, 1:5000) before washing and mounting. Images were acquired on a confocal system (Zeiss LSM 710) using a multi-track configuration.

For analysis of cell morphology, Z-series stacks of confocal images were taken and a single confocal image slice with the largest soma area for individual GFP$^+$ or DsRed$^+$ or GFP$^+$DsRed$^+$ neurons was used for quantification using NIH ImageJ program. For analysis of neuronal positioning, single section confocal images of GFP$^+$ or DsRed$^+$ or GFP$^+$DsRed$^+$ neurons with DAPI staining were used to determine the soma localization within 4 areas defined in **Figure 3A**. For analysis of dendritic development, 3D reconstructions of entire dendritic processes of each newborn neuron were made from Z-series stacks of confocal images. The 2D projection images were traced with NIH ImageJ. All newborn dentate granule neurons with largely intact dendritic trees were analyzed for total dendritic length as described.[11] The measurements did not include corrections for inclinations of dendritic process and therefore represent projected lengths. Sholl analysis for dendritic complexity was carried out by counting the number of dendrites that cross a series of concentric circles at 10 μm intervals from the soma as previously described.[11] For analysis of pS6 levels, primary antibodies against pS6-Ser235/236 (rabbit, 1:1000, Cell Signaling) were used. Effective immunostaining of pS6 required an antigen retrieval protocol as previously described.[18] All analyses were carried out by personnel who were blind to experimental conditions. Multiple animals for each condition were analyzed. Statistical significance was determined by ANOVA or Student's t-test as indicated.

Results

Hyper-activation of Rheb1 is sufficient to activate mTOR signaling in newborn neurons in the adult hippocampus

To examine the function of Rheb1 in newborn granule neurons in the adult hippocampus, we first tested whether Rheb1

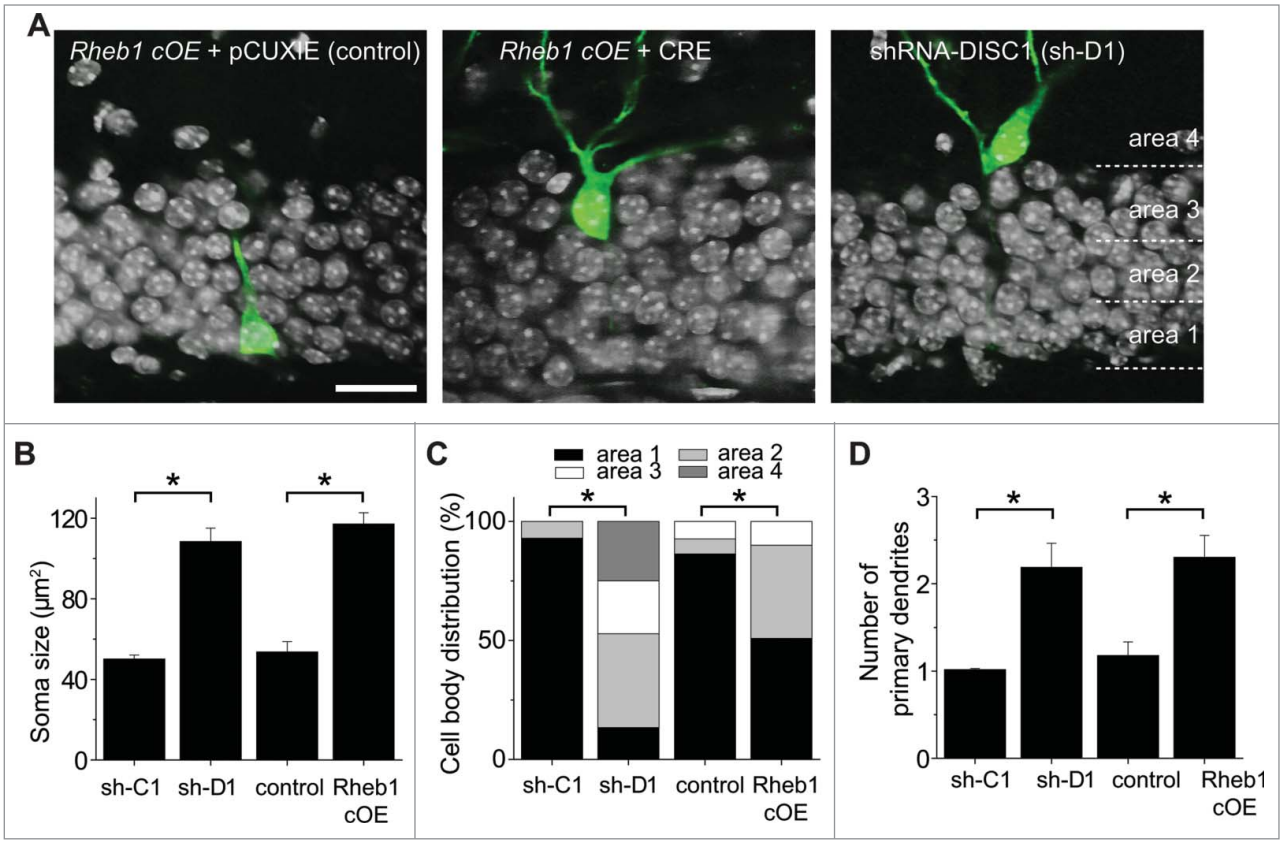

Figure 3. Rheb1 hyper-activation recapitulates DISC1 knockdown-induced cell positioning and morphological defects of newborn neurons in the adult hippocampus. (**A–C**) Soma size and cell body position of newborn neurons after different manipulations at 14 dpi. Shown in (**A**) are sample single-section confocal images to show the soma size and localization within the defined 4 areas in the dentate gyrus. Scale bar: 20 μm. Shown in (**B**) is a summary of soma size. The same groups of cells as in **Figure 2C** were analyzed. Values represent mean + SEM. (*: $p < 0.01$; Student's t-test). Shown in (**C**) is a summary of distribution of GFP^+ cells within each area as defined in (**A**). (**D**) A summary of the number of primary dendrites. The same groups of cells as in **Figure 1C** were analyzed. Values represent mean + SEM. (*: $p < 0.01$; Student's t-test).

hyper-activation specifically in newborn neurons is sufficient to activate the downstream mTOR signaling pathway. In combination with genetically modified mouse conditional lines, we employed a retrovirus-mediated "single-cell" genetic approach for birth-dating and genetic manipulation of proliferating neural progenitors in the adult mouse hippocampus *in vivo*.[28,31] Retroviruses co-expressing Cre recombinase and GFP or those expressing GFP alone were stereotaxically injected into the dentate gyrus of adult conditional *Rheb1* overexpression (cOE) mice, in which a constitutively active form of Rheb1 (S16H) is expressed from the ROSA26 locus in a Cre-dependent fashion[24,32] (**Fig. 1A**). Immunostaining of phosphorylated forms of ribosomal S6 (pS6), an indicator of mTOR activation,[33] and the immature neuronal marker doublecortin (DCX) showed a dramatic increase of pS6 levels in $GFP^+CRE^+DCX^+$ neurons, but not in control $GFP^+CRE^-DCX^+$ neurons at 14 d post viral injection (dpi), compared to unlabeled GFP^-DCX^+ neurons in the same preparation, in which the pS6 level is very low (**Fig. 1B and C**). This result confirms that Cre-mediated constitutively active Rheb1-S16H expression is sufficient to activate mTOR signaling in newborn neurons in the adult hippocampus.

Rheb1 hyper-activation recapitulates DISC1 knockdown-induced developmental defects in newborn neurons in the adult hippocampus

Given that DISC1 knockdown (K_D) leads to increased mTOR signaling in newborn neurons during adult hippocampal neurogenesis,[18] we examined whether Rheb1 hyper-activation in newborn neurons produces similar neurodevelopmental defects as DISC1 K_D. Retroviruses co-expressing Cre recombinase and GFP or GFP alone were stereotaxically injected into the dentate gyrus of *Rheb1* cOE mice, while retroviruses co-expressing GFP with either shRNA against mouse *Disc1* (sh-D1) or scrambled shRNA (sh-C1) were stereotaxically injected into the dentate gyrus of wild type mice (**Fig. 1A and 2A**). Consistent with previous results,[11,18] newborn neurons co-expressing GFP and a previously characterized potent shRNA against mouse *Disc1* (sh-D1) exhibited significantly accelerated dendritic growth at 14 dpi (**Fig. 2B and C**). Interestingly, GFP^+ newborn neurons expressing Rheb1-S16H displayed similarly precocious dendritic developmental defects (**Fig. 2B and C**). Sholl analysis also revealed increased dendritic complexity of newborn neurons with either DISC1 K_D or Rheb1-S16H overexpression (**Fig. 2D**).

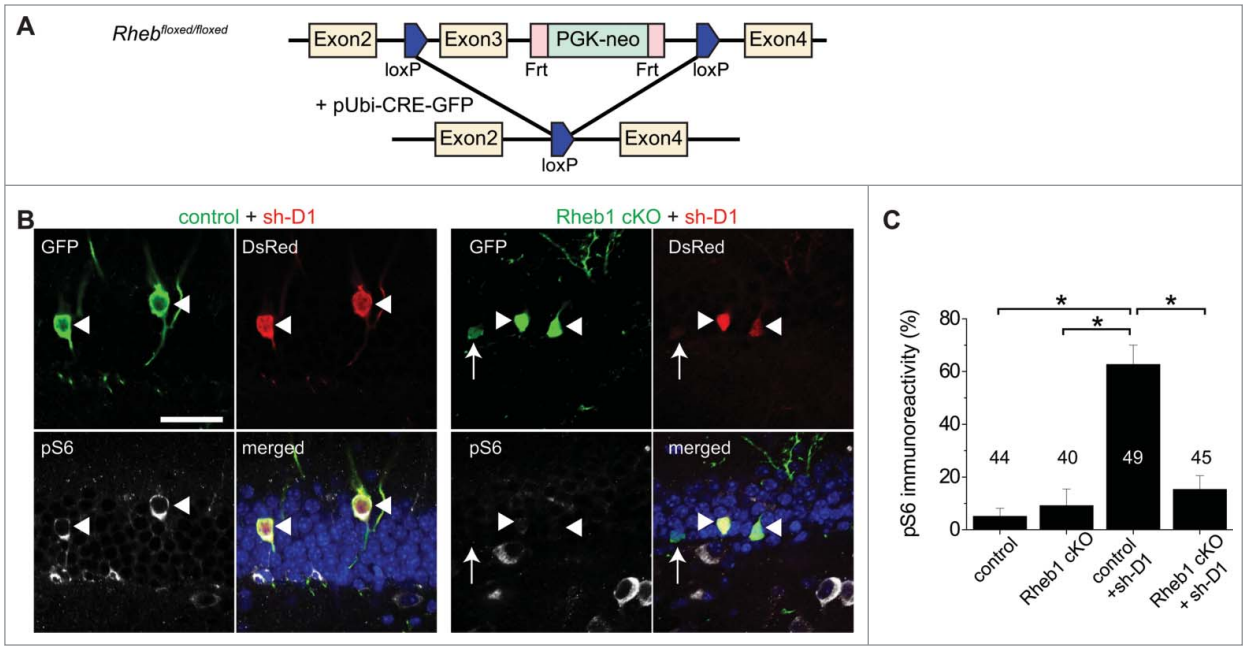

Figure 4. Rheb1 activation is required for DISC1 knockdown-induced increase of mTOR signaling in newborn neurons during adult hippocampal neurogenesis. (**A**) A schematic diagram showing the strategy of knocking out *Rheb1* specifically in proliferating neural progenitors in the adult dentate gyrus. Retroviruses co-expressing GFP and Cre were stereotaxically injected into the dentate gyrus of adult *Rheb1* cKO mice. (**B and C**) *Rheb1* knockout abolished DISC1 K_D-induced increase of pS6-Ser235/236 levels in newborn neurons in the adult *Rheb1* cKO mice. Shown in (**B**) are sample confocal images of immunostaining of GFP and pS6. Arrowheads point to GFP^+DsRed^+ newborn neurons; arrows point to GFP^+ only newborn neurons. Note high pS6 levels in GFP^+ neurons expressing sh-D1 (left panel), but not in those expressing sh-D1 and Cre (right panel). Scale bar: 20 μm. Shown in (**C**) is a summary of quantification of pS6 levels in GFP^+ neurons under different conditions. Numbers associated with the bar graph indicate numbers of GFP^+DsRed^+ neurons examined. Values represent mean + SEM (*: $p < 0.01$; Student's t-test).

We next examined cell positioning and morphology. Consistent with previous studies,[11,18] newborn neurons with DISC1 deficiency exhibited enlarged soma, ectopic primary dendrites, and aberrant cell positioning into the outer granule cell and molecular layers at 14 dpi (**Fig. 3**). GFP^+Cre^+ newborn neurons expressing Rheb1-S16H displayed very similar neuronal developmental defects in soma size and formation of ectopic primary dendrites as DISC1 K_D (**Fig. 3A, B and D**), whereas the neuronal positioning defect appeared to be less severe (**Fig. 3C**). Together, these data suggest that Rheb1 hyper-activation specifically in newborn neurons is sufficient to induce neuronal developmental defects similar to those observed following DISC1 K_D during adult hippocampal neurogenesis.

Rheb1 is required for the DISC1 knockdown-induced increase of mTOR signaling in newborn neurons during adult hippocampal neurogenesis

To investigate whether Rheb1 mediates the increase in mTOR signaling elicited by DISC1 K_D, we used *Rheb1* conditional knockout mice (cKO; **Fig. 4A**). Retroviruses co-expressing GFP and Cre were stereotaxically injected into the dentate gyrus of adult *Rheb1* cKO. No significant change in the immunoreactivity of pS6 was detected for newborn neurons co-expressing GFP and CRE at 14 dpi in the dentate gyrus of adult hippocampus, as the basal level of pS6 was very low (**Fig. 4C**). We further employed a double retroviral manipulation approach for simultaneous *Rheb1* deletion and DISC1 K_D using a mixture of retroviruses co-expressing GFP/CRE and those co-expressing DsRed/sh-D1 in adult *Rheb1* cKO mice. Rheb1 deletion normalized the DISC1 K_D-induced increase of pS6 levels in newborn neurons while control cells with DISC1 K_D alone expressed significantly higher levels of pS6 (**Fig. 4B and C**). This result suggests that endogenous Rheb1 is required for DISC1 K_D-induced over-activation of mTOR signaling in newborn neurons in the adult hippocampus *in vivo*.

Rheb1 is required for DISC1 knockdown-induced neurodevelopmental defects

We next examined whether Rheb1 mediates DISC1 signaling during regulation of neuronal development using *Rheb1* cKO. Unlike Rheb1 hyper-activation, no significant developmental defects were detected for newborn neurons co-expressing GFP and CRE at 14 dpi in the dentate gyrus of adult *Rheb1* cKO mice (**Figs. 5 and 6**), similar to what we found with DISC1 overexpression.[11] For simultaneous *Rheb1* deletion and DISC1 K_D, a mixture of retroviruses co-expressing GFP/CRE and those co-expressing DsRed/sh-D1 were stereotaxically injected into the dentate gyrus of adult *Rheb1* cKO mice. Newborn neurons with only DISC1 K_D exhibited enhanced total dendritic growth and increased dendritic complexity and these DISC1 K_D-induced

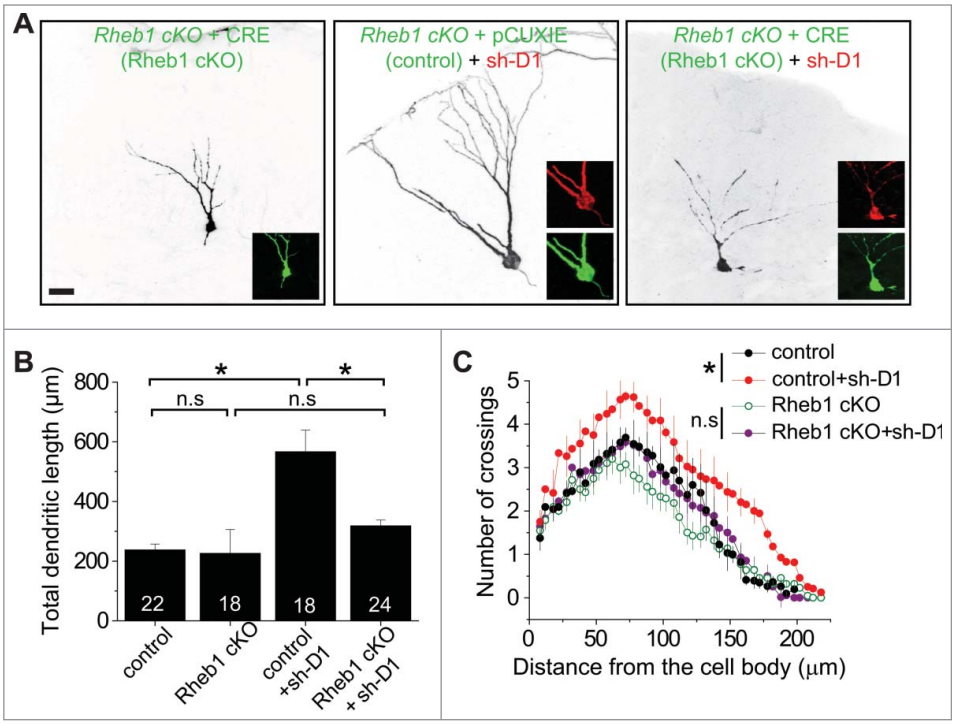

Figure 5. Rheb1 is required for DISC1 knockdown-induced dendritic developmental defects of newborn neurons in the adult hippocampus. (**A–C**) Dendritic length and complexity of newborn neurons at 14 d post retroviral injection (dpi) to co-express GFP alone (control) or co-express GFP and Cre (*Rheb1 cKO*) and DsRed/sh-D1 in adult conditional *Rheb1* KO mice. Shown in (**A**) are sample projection confocal images of newborn neurons under different conditions. Scale bar: 20 μm. Shown in (**B**) is a summary of total dendritic length. Numbers associated with bar graphs indicate total numbers of GFP^+ cells examined from 4 animals for each condition. Values represent mean + SEM. (*: $p < 0.01$; Student's t-test). Shown in (**C**) is a summary of Sholl analysis of dendritic complexity of newborn neurons. The same groups of neurons as in **Figure 5B** were examined. Values represent mean + SEM (*: $p < 0.01$; 2-way ANOVA).

aberrant dendritic phenotypes were rescued by loss of function of *Rheb1* in the same animal (**Fig. 5**). We further analyzed the cell positioning and morphology of DISC1-deficient newborn neurons with or without *Rheb1* expression. *Rheb1* deletion rescued the enlarged soma, abnormal cell positioning and ectopic primary dendrites resulting from DISC1 K_D (**Fig. 6**). Thus, endogenous Rheb1 function is required for DISC1 K_D-induced neuronal developmental defects *in vivo*.

Taken together, our genetic gain- and loss-of-function experiments established Rheb1 as a key mediator of DISC1-dependent regulation of mTOR signaling and neuronal development during adult hippocampal neurogenesis in a cell-autonomous fashion.

Discussion

The scaffold protein DISC1 has many binding partners and thus plays a broad and complex role in neuronal development.[7,34] The mechanism through which DISC1 regulates distinct aspects of neuronal development *in vivo* is not well understood. The mTOR signaling pathway, as a major pathway controlling cellular metabolic homeostasis, has been proposed to function as a signaling hub to integrate external stimuli for the regulation of many biological processes, including neural development and neuropsychiatric diseases.[35] However, due to the complexity of this pathway, with multiple upstream regulators and downstream effectors,[33] it has been difficult to dissect the components that regulate neuronal development. Although previous results have demonstrated that systemic administration of the mTOR antagonist rapamycin could ameliorate effects of DISC1 knockdown in neuronal development,[17-19] it was not known whether the relevant changes in mTOR signaling were operating in a cell-autonomous manner. Using *in vivo* "single-cell" genetic analyses in conditional knockout and conditional overexpression mouse models to selectively manipulate Rheb1 in adult-born neurons, we demonstrate here a cell-autonomous role of Rheb1 in mediating DISC1 regulation of neuronal development.

Adult hippocampal neurogenesis is an exceptionally advantageous *in vivo* cellular model system for investigating functions and signaling pathways of risk genes for mental disorders in neuronal development.[36-38] It recapitulates the complete process of embryonic neuronal development, including proliferation, fate determination, migration, axon/dendrite development and synaptic integration, with a prolonged time course, allowing for high resolution analysis of each stage of neuronal development and its associated molecular events. In this study we have identified Rheb1 as a key mediator linking DISC1 to the mTOR pathway in regulating neuronal development. Rheb1 was originally identified by differential cloning as a small GTPase in the Ras family that is induced by synaptic activity in dentate granule cells in the context of seizures and long-term potentiation.[21] The physiological role of Rheb1 *in vivo* has only recently been a target of investigation because of the availability of newly developed conditional genetic mouse models, as a direct knockout is embryonic lethal. For example, specific deletion of Rheb1 in the oligodendrocyte precursor cells using Olig1/2-Cre, or in the brain using nestin-Cre, results in defects in oligodendrocyte precursor proliferation and myelination during brain development, respectively, and both processes are dependent on signaling through mTOR complex 1, but not TORC2.[24,32] In the current study, combining *in vivo* single-cell manipulation using a retrovirus to express Cre and conditional Rheb1 overexpression mouse lines, we provided evidence that

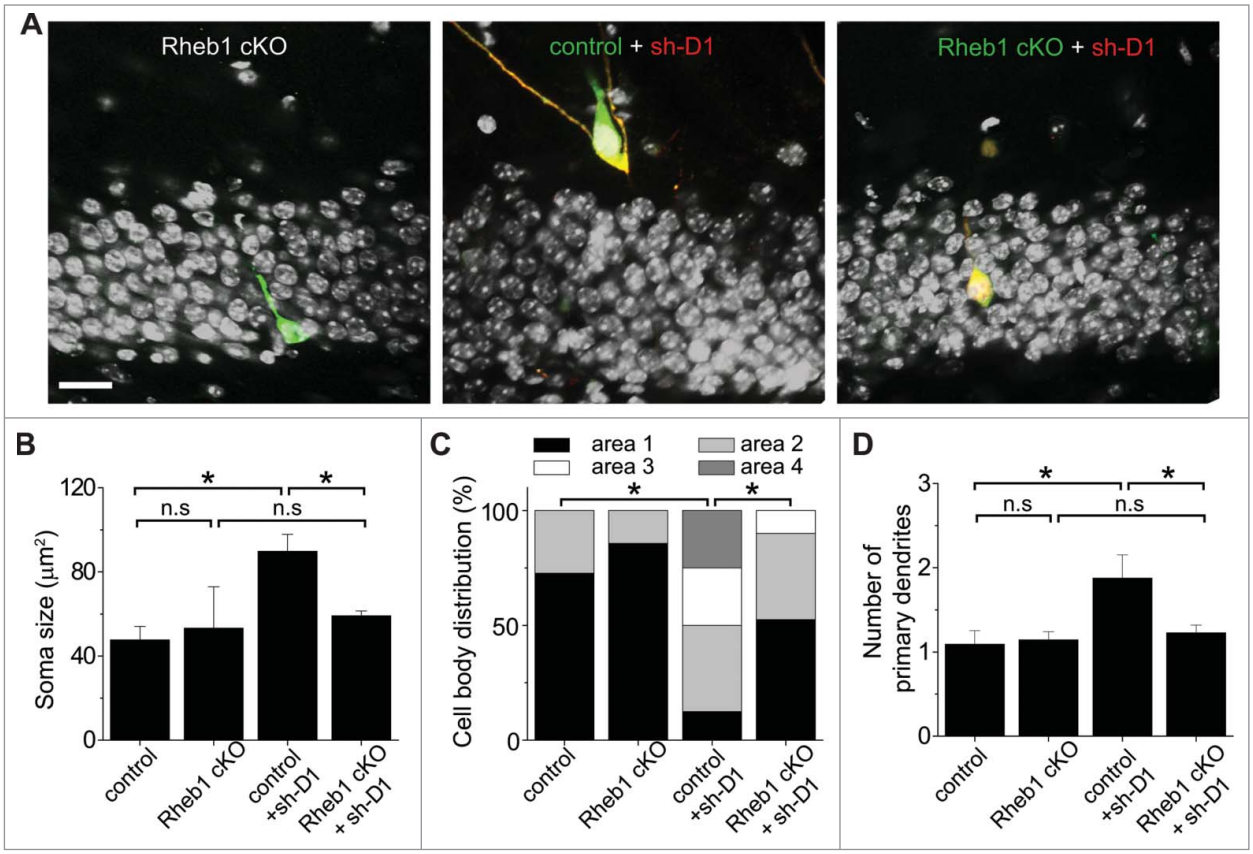

Figure 6. Rheb1 function is required for DISC1 knockdown-induced cell positioning and morphological defects of newborn neurons in the adult hippocampus. (**A–C**) Soma size and cell body position of newborn neurons after different manipulations at 14 dpi. Shown in (**A**) are sample single-section confocal images to show soma size and localization of newborn neurons within the defined 4 areas in the dentate gyrus. Scale bar: 20 μm. Shown in (**B**) is a summary of soma size. The same groups of cells as in **Figure 5B** were analyzed. Values represent mean + SEM. (*: $p < 0.01$; Student's t-test). Shown in (**C**) is a summary of the distribution of GFP^+ cells within each area as defined in **Figure 3A**. (**D**) A summary of the number of primary dendrites. The same groups of cells as in **Figure 5B** were analyzed. Values represent mean + SEM. (*: $p < 0.01$; Student's t-test).

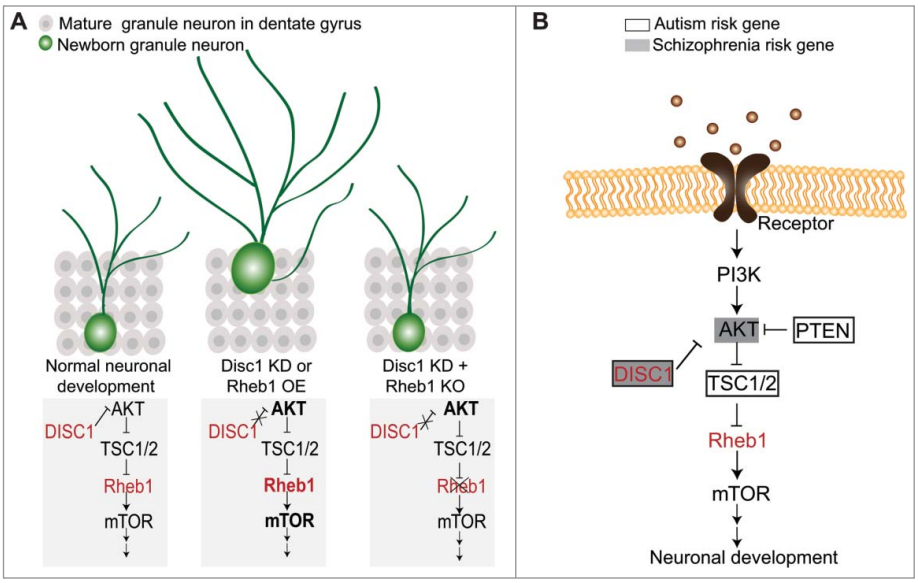

Figure 7. A summary model of DISC1 signaling mediated by Rheb1 in regulating neuronal development during adult hippocampal neurogenesis.

phenotypes resulting from over-activation of Rheb1 closely resemble those seen with DISC1 knockdown. More importantly, we found that deletion of Rheb1 normalizes mTOR activity and rescues cellular phenotypes induced by DISC1 knockdown. Thus, Rheb1 is both required and sufficient to mediate DISC1-dependent regulation of neural development in newborn neurons in the adult hippocampus (**Fig. 7A**).

In addition to its putative relevance in neurodevelopmental disorders, such as ASD and SCZ, Rheb1 is a direct target of TSC1/2, the protein complex encoded by the *TSC1* and *TSC2* genes. Heterozygous mutations in *TSC1* or *TSC2* are the cause of tuberous sclerosis, a neurocutaneous disorder with a high incidence of brain involvement, intellectual disability and co-morbid diagnoses

of ASD.[39] Independently, genetic association studies have identified *TSC1* and *TSC2* as risk genes for ASD.[40] Our results implicate TSC1/2 in the Rheb1-mediated effect of DISC1 knockdown in adult neurogenesis, potentially linking another ASD risk gene to the DISC1 signaling cascade (**Fig. 7B**), which may subsequently reveal common targets for intervention, diagnostic markers and treatment of multiple mental disorders.

Furthermore, the signaling pathway we have delineated in the development of adult newborn neurons may have direct relevance to SCZ, as disruptions in post-natal neurogenesis have been implicated in the pathogenesis of this disorder. Postmortem brains from patients with SCZ show a reduced number of proliferating neural progenitors,[41] immaturity of dentate gyrus granule neurons[42] and diminished volume of the hippocampus.[43] While it remains an interesting idea that disturbances in adult hippocampal neurogenesis may contribute to psychiatric disorders, the signaling mechanism discovered here provides a foundation to understand how multiple risk genes functionally converge into common pathways that may be responsible for the etiology of psychiatric disorders and could be potential targets for therapeutic interventions.

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

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