Changes in ADAR1 activity during *Plasmodium* infection contribute to protection from malaria.

Jaclyn Quin[1], Eli Kopel[2], Riem Gawish[3], Michelle Eidelman[2], Dragana Vukić[1,4], Pavla Linhartová [1], Janka Melicherová[1,4], Ketty Sinigaglia[1], Sajjad Ghodrati[5], Charles Arama[6], Issa Nebie[7], Marita Troye-Blomberg[8], Eva Sverremark-Ekström[8], Sylvia Knapp[3], David Modry[5,9,10], Ann-Kristin Östlund-Farrants[8], Erez Levanon[2], Liam P. Keegan[1], and Mary A. O'Connell[1]*.

[1]CEITEC Masaryk University; Brno, Czech Republic. [2]The Mina and Everard Goodman Faculty of Life Sciences, Bar-Ilan University; Ramat Gan, Israel. [3]Research Division Infection Biology, Department of Medicine I, Medical University of Vienna; Vienna, Austria. [4]National Centre for Biomolecular Research, Faculty of Science, Masaryk University; Brno, Czech Republic. [5]Department of Botany and Zoology, Faculty of Science, Masaryk University; Brno, Czech Republic. [6]Malaria Research and Training Centre, Department of Epidemiology of Parasitic Diseases, International Center of Excellence in Research, University of Sciences, Technique and Technology of Bamako; Bamako, Mali. [7]Groupe de Recherche Action en Santé; Ouagadougou, Burkina Faso. [8]Department of Molecular Biosciences, The Wenner-Gren Institute, Stockholm University; Stockholm, Sweden. [9]Department of Veterinary Sciences FAPPZ and CINeZ, Czech University of Life Sciences; Prague, Czech Republic. [10]Parasitological Institute of CAS, Biology Center; České Budějovice, Czech Republic.

*Corresponding author. Email: mary.oconnell@ceitec.muni.cz

Summary.

Adenosine deaminase acting on RNA 1 (ADAR1) suppresses the activation of multiple antiviral immune response pathways. Here, we investigate the role of ADAR1 during infection with the *Plasmodium* parasite, which causes malaria and is responsible for over almost a half million childhood deaths every year. Reduced activity of ADAR1 during *Plasmodium* infection is associated with populations protected from clinical malaria. In animal models, *Adar*$^{+/-}$ mice are protected from *P. yoelii* parasitemia, via a previously unreported pathway. These mice display elevated Type-I IFN responses and CD8^{+} T cell activation, but no detrimental immune responses. Our results suggest that a decrease in the levels of ADAR1 occurs during infection and can drive both innate and adaptive immune responses, and this presents a previously unrecognized opportunity for targeting ADAR1 in diverse infectious diseases.

Main Text.

ADAR1 converts adenosine (A) to inosine (I) in double-stranded RNA (dsRNA) by hydrolytic deamination, a process called A-to-I RNA editing. A-to-I RNA editing by ADAR1 is widespread in human RNA, occurring at millions of sites in stretches of dsRNA which form within the majority of transcripts (*1, 2*). An essential function of ADAR1 is to prevent harmful activation of innate immunity. Particularly, the classical role of ADAR1 is to prevent 'self'-dsRNAs from activating immune sensors that detect viral and other 'non-self'-dsRNAs in the cytoplasm. Such innate immune sensors include the RIG-I like receptors (RLRs) (*3-5*), protein kinase R (PKR) (*6-12*), oligoadenylate synthetase (OAS)–ribonuclease (RNase) L (*13-16*), and Z-D/RNA binding protein 1 (ZBP1) (*17-22*). When activated by dsRNAs, these pathways drive antiviral innate immune responses, including expression of Type-I interferons (IFNs) and proinflammatory cytokines, inhibition of translation, cleavage of cellular RNAs, and/or activation of cell death pathways (*23-25*) (Fig. 1A). Mutations in ADAR1 have been found to cause autoimmune disorders which are characterized by aberrant Type-I IFN responses and chronic inflammation (*26*). On the other hand, overexpression of ADAR1 and increased A-to-I RNA editing is common across different cancer types (*27, 28*), and targeting ADAR1 can result in cancer cell lethality by driving anti-tumor immune responses (*29*).

Relatively little is known about the regulation of ADAR1 during infection, and how this impacts disease outcomes (*30, 31*). ADAR1 has two isoforms: ADAR1p110, which localizes

predominantly within the nucleus; and ADAR1p150, which localizes in both the nucleus and cytoplasm, and is induced by IFN(*32, 33*). Significant spatiotemporal changes occur in levels of A-to-I RNA editing by ADAR1 (*34-36*), and a number of reports illustrate that ADAR1 activity is sensitively and dynamically controlled by diverse mechanisms (*37*). However, our understanding of pathways of regulation of ADAR1 activity remains largely incomplete. During viral infections, ADAR1 activity can suppress antiviral immune response pathways. For example, knock-down of ADAR1 increases IFN and/or PKR signaling in infected cell line models (*7, 30, 38-42*). However, ADAR1 can also directly edit virus dsRNA, which can be either pro-viral or antiviral in different contexts (*43*). Despite the abundance of evidence that ADAR1 can impact viral infections in diverse ways, its role in other types of infections has been almost entirely unexplored(*44, 45*).

Malaria is caused by infection with *Plasmodium*, which are single-celled eukaryotic protozoan parasites. Here, we identify changes in A-to-I RNA editing by ADAR1 in the Fulani, a West African ethnic group protected from malaria (*46*). This led us to investigate whether ADAR1 activity impacts malaria infection. In different human models, we find A-to-I editing of endogenous 'self' RNA is altered following malaria infection, and lower A-to-I editing is associated with protection from the disease. Strikingly, *Adar* (encoding Adar1) heterozygous mutant mice are significantly protected from blood stage parasitemia during infection with rodent *P. yoelii* malaria. *Adar* heterozygous mutant mice display activation of Type-I IFN and $CD8^+$ T-cell responses, at a relatively controlled level at baseline, and these are further activated during *P. yoelii* infection. Therefore, our data suggests that ADAR1 impacts infection with *Plasmodium* protozoan parasite, and that reduction of ADAR1 activity during malaria contributes to protection from the disease.

A reduced level of A-to-I RNA editing by ADAR1 following *Plasmodium* infection is associated with protection from malaria. The Fulani ethnic group of the Sahel region of Africa are protected from *P. falciparum* malaria(*46*). Despite similar exposure as other sympatric ethnic groups, the Fulani have heightened innate and adaptive immune responses to *P. falciparum,* lower parasite densities in infected individuals, and fewer symptomatic cases of malaria (*47*). However, the underlying basis of their protection is unknown. We previously performed RNA-sequencing analysis of $CD14^+$ peripheral blood mononuclear cells (PBMCs) isolated from the Fulani during the *P. falciparum* infection season (*48*). Levels of A-to-I editing by ADAR1 can be readily

determined from RNA-sequencing data using established methods, as inosines (I) are recognised as guanosines (G) by reverse transcriptase during cDNA synthesis, creating A-G mismatches during sequence alignment (*49, 50*). Here, we show that during *P. falciparum* infection, global levels of A-to-I editing measure by analysis of the Hyperediting Index in $CD14^+$ PBMCs are reduced in the Fulani compared to a sympatric ethnic group (Fig. 1B). This indicates that ADAR1 activity levels are differently modulated in susceptible versus protected individuals during malaria. We extended our analysis to publicly available RNA-sequencing data from human models of *Plasmodium* infection. Controlled human malaria infection (CHMI) with *P. falciparum* in malaria-naïve individuals results in increased A-to-I editing levels in whole blood from pre- to post-challenge (*51*) (Fig. 1C). In contrast, individuals who have undergone immunization with attenuated *P. falciparum* sporozoites (*Pf*SPZ) have reduced A-to-I editing levels in whole blood following the final *Pf*SPZ infection (*52*) (Fig. 1D). Individuals who have undergone immunization with attenuated *P. vivax* sporozoites (*Pv*RAS) also have reduced A-to-I editing levels in PBMCs both following the final *Pv*RAS infection and during subsequent challenge with *P. vivax* (*53*) (Fig. 1E). Further analysis of a cohort of young individuals living in a *P. falciparum* endemic region as they acquire natural protection from malaria, suggests that exposure to *Plasmodium* results in reduced levels of global editing by ADAR1p110 in protected individuals, and increased rates of editing predominantly by IFN-inducible ADARp150 in susceptible individuals(*51*) (fig. S1). Overall, reduced levels of global A-to-I editing by ADAR1 during *Plasmodium* infection are associated with protection from malaria.

***Adar*$^{+/-}$ heterozygous mutant mice have a benign Type I IFN phenotype and are protected from *P. yoelii* parasitemia.** In order to determine if reduced ADAR1 activity can contribute to protection during malaria, we performed infection of *Adar* heterozygous mutant mice with *P. yoelii* 17XNL non-lethal rodent malaria. *Adar*$^{-/-}$ homozygous null mice are embryonic lethal at day E11.5-E12.5, associated with overproduction of Type-I IFN, loss of embryonic liver hematopoietic cells, liver disintegration, and widespread apoptosis (*6, 54*). The embryonic lethal phenotype can be rescued to several days after birth in double mutants lacking *Adar*$^{-/-}$ *Ifih1*$^{-/-}$ encoding melanoma differentiation-associated protein 5 (Mda5) and in *Adar*$^{-/-}$ *Mavs*$^{-/-}$ (mitochondrial antiviral-signaling *protein*) double mutants, which prevent activation of the RLR/MAVS pathway, but these mice do not survive to adulthood (*3-5*). Here, we have utilised *Adar*$^{+/-}$ heterozygous mutant mice, which are born at Mendelian ratios and are phenotypically normal (*6, 54, 55*). We have previously

shown that mouse embryonic fibroblast (MEF) cultures from these mice have heightened Type-I IFN responses following stimulation with Poly(I:C) dsRNA (*3*). We performed RNA-sequencing analysis of whole blood of adult wild-type and *Adar*$^{+/-}$ mice. Only a small number of genes are significantly differentially expressed (DE) in *Adar*$^{+/-}$ mice (27 genes, Adjusted p-value <0.05, Log2FoldChange >1) (Fig. 2A&B, and table S1). Gene ontology (GO) enrichment analysis shows that these are significantly enriched for genes involved in cellular response to IFNs, antiviral response, and innate immunity (table S1). All DE genes are IFN-stimulated genes (ISGs) (27/27 genes; Interferome database (*56*)). Analysis of cytokine levels in plasma collected from peripheral blood of adult wild-type and *Adar*$^{+/-}$ mice shows only a small but significant increase in CXCL10, a sensitive marker of Type-I IFN activation (Fig. 2C, and fig. S2). Immunoblot analysis of the livers of adult mice shows slightly increased protein levels of ISGs, including RIG-I and PKR, in *Adar*$^{+/-}$ mice (Fig. 2D). *Adar*$^{+/-}$ mice maintain a normal body weight and blood cell composition (fig. S3). Collectively, our analyses indicate that *Adar*$^{+/-}$ mice have mildly increased levels of Type-I IFN responses. This increase is controlled, and does not cause any apparent deleterious consequences during a normal lifespan. We infected age-matched pairs of wild-type and *Adar*$^{+/-}$ mice with 10^5 *P. yoelii* 17XNL:PyGFP infected red blood cells (iRBCs) via tail vein injection, and monitored levels of parasitemia until self-resolution (approximately 4 weeks). The maximum *P. yoelii* parasitemia was significantly lower in *Adar*$^{+/-}$ mice. *P. yoelii* parasitemia began to diverge after 1 week of infection, and remained significantly lower across the course of infection in *Adar*$^{+/-}$ mice (Fig. 2E). Therefore, reduced levels of Adar1 protect against *P. yoelii* parasitemia.

***Adar*$^{+/-}$ mice are protected from *P. yoelii* parasitemia independently of the RLR pathway.** To investigate the mechanisms underlying the protection of *Adar*$^{+/-}$ mice from *P. yoelii* parasitemia, we performed RNA-sequencing analysis of whole blood from adult wild-type and *Adar*$^{+/-}$ mice at 7 days following *P. yoelii* infection. At day 7, there are no significant differences in parasitemia between wild-type and *Adar*$^{+/-}$ mice (fig. S4). We observed very few significant differences in gene expression, with only 6 DE genes in *P. yoelii*-infected *Adar*$^{+/-}$ mice compared to *P. yoelii*-infected wild-type mice (Adjusted p-value <0.05, Log2FoldChange >1) (Fig. 3A&B, and table S1). There were a large number of DE genes when comparing infected mice to uninfected mice for both genotypes (wild-type day 7 vs wild-type uninfected, 1420 DE genes; *Adar*$^{+/-}$ day 7 vs *Adar*$^{+/-}$ uninfected, 1796 DE genes (Adjusted p-value <0.05, Log2FoldChange >1)), with a significant overlap in DE genes between wild-type and *Adar*$^{+/-}$ mice (1004 genes, or 70% of wild-

type DE genes, and 56% *Adar*$^{+/-}$ DE genes) (fig. S5). Together, this suggests that wild-type and *Adar*$^{+/-}$ mice respond in a generally similar manner to *P. yoelii* infection but responses are higher in *Adar*$^{+/-}$ mice. The DE gene *Treml1* is specifically expressed in platelets and megakaryocytes, and used as a sensitive marker of platelet activation (*57*). We performed analysis of platelets in peripheral blood of uninfected mice, and observed slightly increased platelet counts and plateletcrit in *Adar*$^{+/-}$ compared to wild-type mice, providing phenotypic validation of our RNA-sequencing results (fig. S6).

As outlined above, Adar1 is known to prevent activation of dsRNA receptors, including RLRs, PKR, OAS-RNASeL and ZBP1. Of these, only the RLR pathway has been implicated in the immune response to malaria. Type-I IFN signalling is activated by *Plasmodium* infection early in liver stage in an Mda5- and Mavs-dependent manner, and mice that are deficient for components of this pathway have reduced ability to control both liver- and blood-stage infection (*58, 59*). Therefore, we investigated the hypothesis that reduced levels of A-to-I editing of dsRNA in *Adar*$^{+/-}$ mice activate the RLR pathway to confer protection from *P. yoelii* parasitemia. At baseline, *Adar*$^{+/-}$ mice do not appear to have significantly reduced levels of A-to-I editing (Fig. 3C). During *P. yoelii* infection both wild-type and *Adar*$^{+/-}$mice display a trend for reduced levels of global A-to-I editing. In contrast, A-to-I editing of 3' UTRs of mRNAs, which are enriched in Adar1p150 target sites, are significantly increased in *Adar*$^{+/-}$ mice (Fig. 3C). These results reflect those reported for human models of malaria infection above. We also confirmed that *P. yoelii* RNA is not targeted for A-to-I RNA editing by Adar1 during infection of the host (fig. S7). Levels of some cytokines, including CXCL10 and TNF-α, increase more significantly in *Adar*$^{+/-}$ mice than in wild-type mice following *P. yoelii* infection (Fig. 3D, and fig. S8). However, the increase is slight and does not indicate a strong pro-inflammatory phenotype in the *Adar*$^{+/-}$ mice. Finally, we infected RLR pathway mutant *Mavs*$^{-/-}$ and *Mavs*$^{-/-}$*Adar*$^{+/-}$ mice with *P. yoelii,* under the same conditions as described above. *P. yoelii* parasitemia is significantly lower after 14 days of infection in *Mavs*$^{-/-}$*Adar*$^{+/-}$ compared to *Mavs*$^{-/-}$ mice. We could not decisively compare the protection between all four genotypes (wt, *Adar*$^{+/-}$, *Mavs*$^{-/-}$, and *Mavs*$^{-/-}$*Adar*$^{+/-}$) as these mice were not sibling pairs. However, the level of protection between *Adar*$^{+/-}$ and *Mavs*$^{-/-}$*Adar*$^{+/-}$ mice is equivalent (Fig. 3E). Therefore, reduced levels of Adar1 can contribute to protection against *P. yoelii* parasitemia in an RLR pathway-independent manner.

***Adar*$^{+/-}$ mice display T cell phenotypes associated with protection from malaria.** Our current understanding of the key immune responses that contribute to establishing resistance to malaria suggest that clinical immunity (for example, after experiencing multiple *Plasmodium* infections) is acquired mainly through memory CD4$^+$ T cells, which produce pro-inflammatory responses and contribute to activation of B cells and production of antibodies, and also through memory CD8$^+$ T cells, which actively kill infected cells (*60*). On the other hand, sterile immunity (for example, following effective vaccination) appears to be acquired mainly through blocking of infection through high antibody titres, and killing of infected cells by memory CD8$^+$ T cells in the liver stage (*61, 62*). Prior studies have shown that hematopoietic cell types are particularly affected by Adar1 loss (*5, 54, 55, 63, 64*). *Adar* is essential for erythropoiesis (*65*), T cell development (*66*), and B cell development (*67, 68*). On the other hand, *ADAR* has been identified as a 'top hit' in loss-of-function CRISPR screens to both restore tumor cell line responses to CD8$^+$ T cell dependent immune checkpoint blockade therapy (*69*), and to induce anti-tumor CD8$^+$ T cell killing (*11, 70*). Therefore, we immunologically characterised *Adar*$^{+/-}$ mice to determine whether reduced Adar1 levels hinder or promote malaria protective immune responses.

Analysis of the thymus and bone marrow indicates that normal T cell and B cell development is preserved in *Adar*$^{+/-}$ heterozygous mice (fig. S9). However, in peripheral blood, while we find the total leukocyte numbers unaffected, *Adar*$^{+/-}$ mice present with a different cellular composition (fig. S10). The abundance of CD8$^+$ T cells is increased, and intriguingly, there are fewer naïve and more memory CD8$^+$ T cells in uninfected *Adar*$^{+/-}$ mice (Fig. 4A). The spleen is central to the immune response to blood stage *Plasmodium* infection, acting as a significant site of infected red blood cells (iRBC) sequestration (*71, 72*), removing iRBCs from circulation, and acting as a secondary lymphoid organ to activate immune responses (*73*). In the spleen of uninfected *Adar*$^{+/-}$ mice, leukocyte counts are slightly elevated, including for B cells and T cells (fig. S10). Similarly to blood, we specifically find CD8$^+$ T cells to be elevated, with fewer naïve, and more effector and memory CD8$^+$ T cells in uninfected *Adar*$^{+/-}$ mice (Fig. 4B). During infection with *P. yoelii* (day 5), at which stage parasitemia remains the same between the two genotypes, the blood cell composition is indistinguishable between wild-type and *Adar*$^{+/-}$ mice (data not shown). However, while all mice develop profound splenomegaly, spleen weights are significantly lower in *Adar*$^{+/-}$ mice (Fig. 4C). These differences are not caused by leukocytes, as total splenic leukocyte numbers remain higher in infected *Adar*$^{+/-}$ mice (fig. S10). During infection with *P. yoelii*, *Adar*$^{+/-}$ mice

present with increased numbers of both $CD4^+$ and $CD8^+$ T cells, including naïve, effector, and memory T cell populations (Fig. 4D). This increase occurs independently of the RLR pathway, as *Mavs*$^{-/-}$ *Adar*$^{+/-}$ mice also have significantly increased $CD4^+$ and $CD8^+$ T cell populations (fig. S11). Together, reduced levels of Adar1 can drive protective T cell responses during *Plasmodium* infection, by an as yet unidentified mechanism.

Discussion. Here we show that targeting ADAR1 can offer protection from *Plasmodium* parasites. These parasites are biologically far more complex organisms than the viruses that have been the focus of past studies on the role of ADAR1 in infection. Our findings indicate that ADAR1 may impact a much broader range of infectious diseases types, including malaria. Key responses for establishing sterile protection from malaria can be induced by targeting ADAR1, including Type-I IFN signaling (*11, 74*), and cytotoxic $CD8^+$ T-cell killing(*11, 69, 70*) (*61, 62*). In our model, reduced activity of ADAR1 does not impact the pathogen directly, but rather independently activates host immune responses to offer protection from disease.

ADAR1 has numerous emerging roles in coordinating the immune response. The regulation of antiviral innate immune response pathways via cytosolic dsRNA sensors RLRs, PKR, OAS-RNAse L and ZBP1 is well established. More recent studies, for example, report that ADAR1 impacts antigen presenting cell (APC) activation and signaling to T cells (*75-77*), as well as T cell dependent antibody responses in activated B cells (*78*). However, to date this is the first report in an animal model of protection against infection with microorganisms due to a decrease in Adar1 levels. This result was unexpected and opens new opportunities for exploration.

ADAR1 has been identified as a 'top hit' in multiple genome wide studies to identify targets that sensitize cancers to the immune response (*69, 70, 79-81*). These results have motivated a race for ADAR1 inhibitors, and a number of candidates are currently in development (*82-84*). Considering the multitude of dynamic pathways through which ADAR1 may influence infection, there are advantages in performing studies in live animals so that the entire host response to the disease can be considered. We show here that *Adar*$^{+/-}$ mice are a robust model to investigate the impact of Adar1 on infection. Despite having only mild inflammation and a normal life span, they have significant protection from malaria. Promisingly, this suggests that a just a 50% decrease in *Adar* can be beneficial, and that there is ample therapeutic window for targeting ADAR1. In this context,

it is timely to investigate the role of ADAR1 in different infections, and to identify the mechanisms through which it confers protection.

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Author contributions.

Conceptualization: JQ, MOC

Methodology: JQ, MOC, EYL, RG

Investigation: JQ, EK, RG, ME, DV, PL, JM, KS, SG, IN, CA

Visualization: JQ

Funding acquisition: JQ, MOC

Project administration: JQ, MOC

Supervision: JQ, IN, MTB, ESE, SK, DM, AOF, EYL, LK, MOC

Writing – original draft: JQ

Writing – review & editing: EK, RG, ME, DV, PL, JM, CA, ESE, AOF, EYL, LP, MOC

Declarations.

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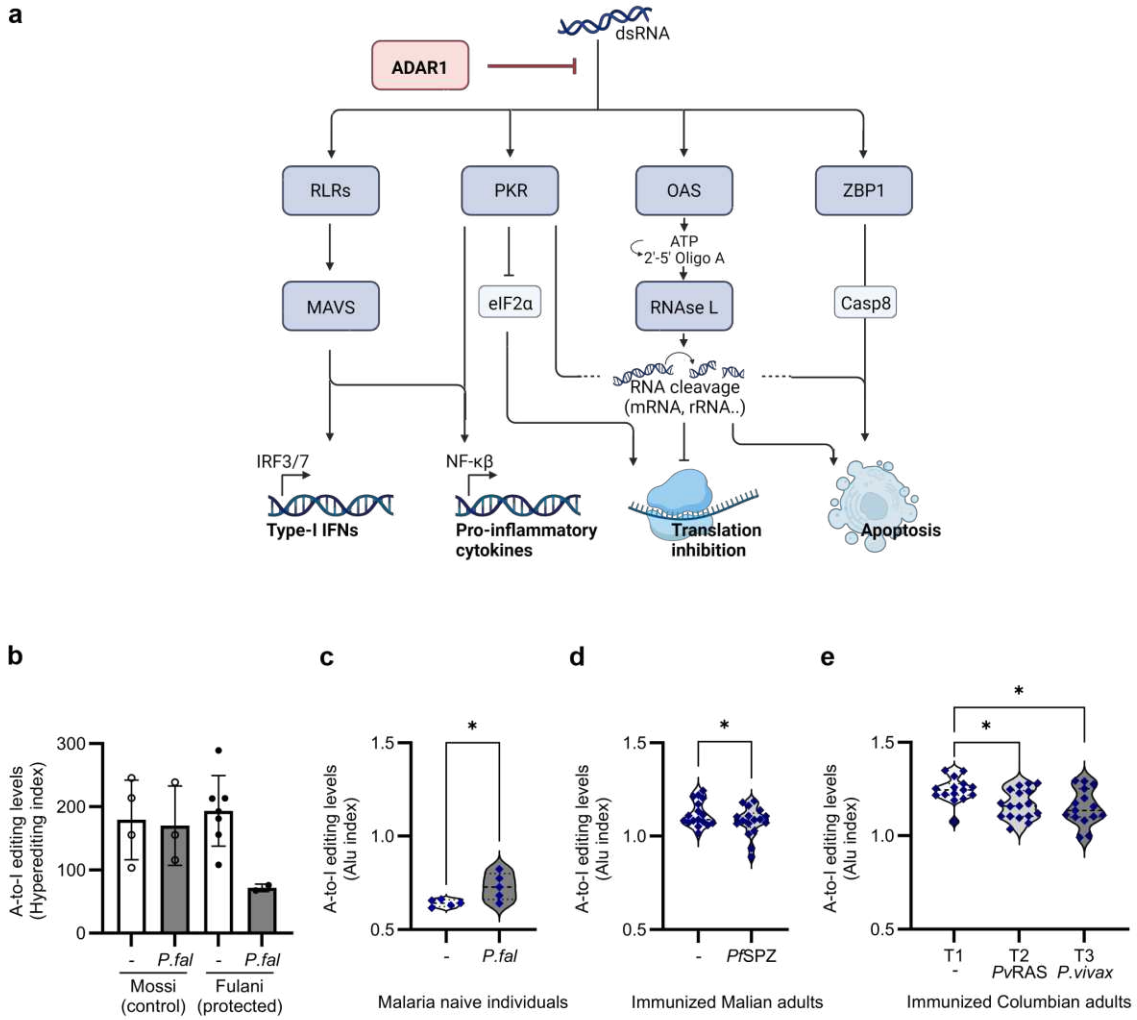


Fig. 1. A reduced level of A-to-I RNA editing by ADAR1 following *Plasmodium* infection is associated with protection from malaria. (**A)** ADAR1 prevents activation of cytopsolic dsRNA receptors and downstream anti-viral immune responses. (**B-E)** Global A-to-I RNA editing levels in human blood determined by analyses of RNA-sequencing data. (**B)** $CD14^+$ PBMCs of protected Fulani and sympatric Mossi individuals uninfected or infected with *P. falciparum*. Hyperediting index (total edited sites/uniquely mapped reads). (Mossi – *P. fal* (n=4 179.4 +/-62.9 s.d.); Mossi + *P. fal* (n=3 170.1 +/-62.7 s.d.); Fulani – *P. fal* (n=7 193.4 +/-55.9 s.d.); Fulani + *P. fal* (n=2 72.0 +/-5.9 s.d.). (**C)** Whole blood of malaria-naive Dutch adults following CHMI with *P. falciparum* (GSE50957). Alu editing index (n=5. Pre-challenge vs post-challenge *P=0.0477 paired t-test). (**D)** Whole blood of Malian adults after five rounds of immunization with attenuated *P. falciparum* sporozoites (Sanaria PfSPZ) every 4 weeks (GSE86308). Alu editing index (n=17. Pre-vaccination (day -7) vs post-vaccination (day 143) *P=0.0203 paired t-test). (**E)** PBMCs isolated from malaria-naive Columbian volunteers, at baseline (T1), after seven rounds of immunization with *attenuated P. vivax* sporozoites (PvRAS) every 8 weeks (T2), and at first diagnosis of infection after a subsequent *P. vivax* challenge (T3) (GSE85263). Alu editing index. (n=15-17. Mean +/- s.d. T1 vs T2 *P=0.0291, T1 vs T3 *P=0.0100 ordinary one-way ANOVA with Dunnett's test for multiple comparisons).

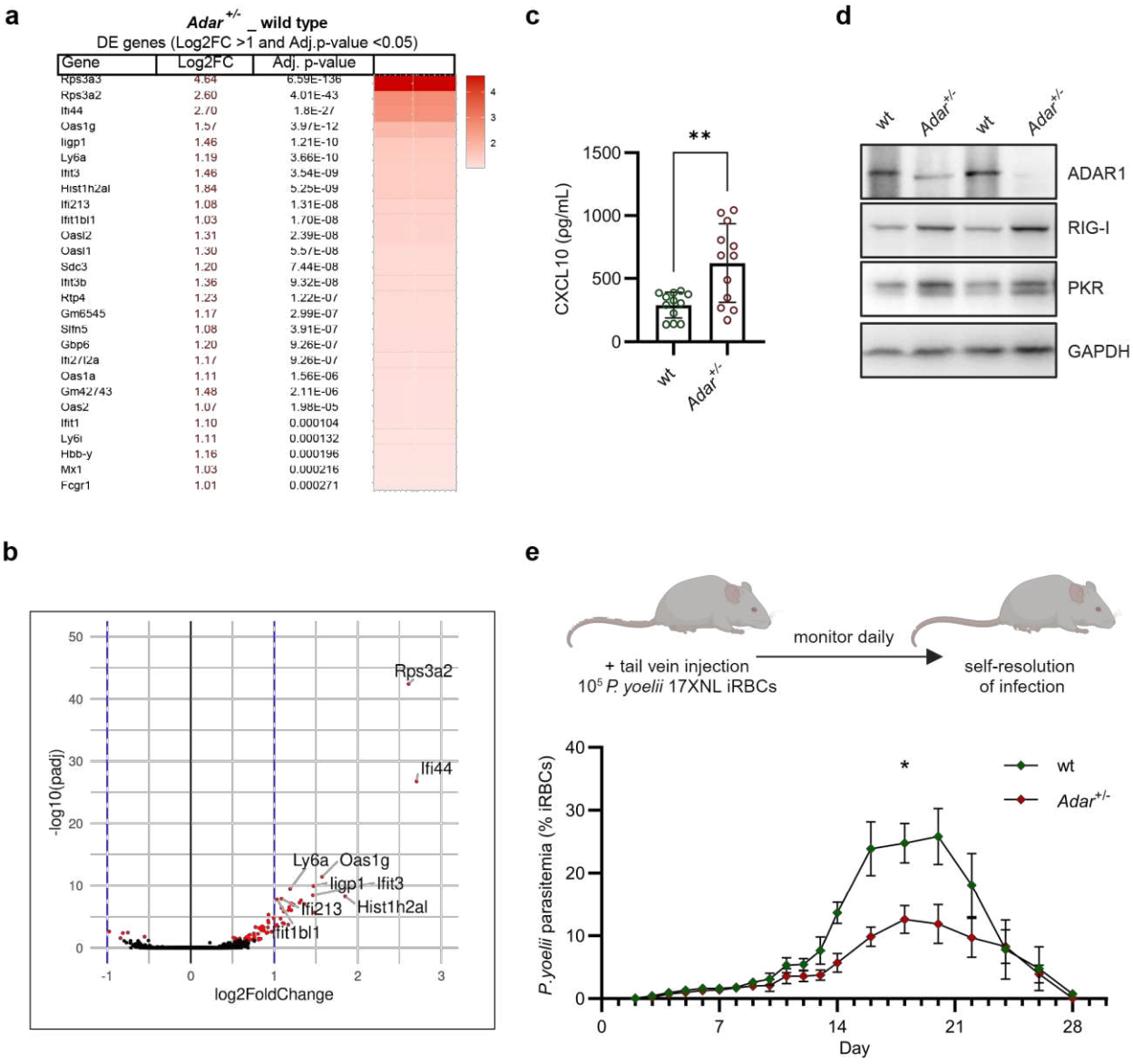


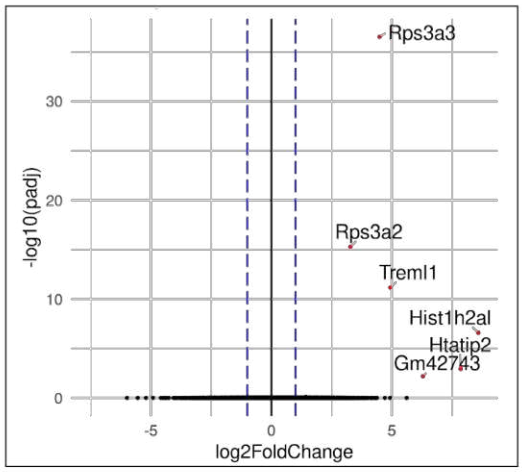
Fig. 2. *Adar*$^{+/-}$ heterozygous mutant mice have a benign Type I IFN phenotype and are protected from *P. yoelii* parasitemia. (A-B) RNA-sequencing analysis of whole blood of adult wild-type and *Adar*$^{+/-}$ mice (n=4 age-matched pairs). (**A)** Table of differentially expressed (DE) genes (27 genes, Adjusted p-value <0.05, Log2 FoldChange>1). (**B)** Volcano plot of DE genes. Significantly DE genes are shown in red (65 genes, Adjusted p-value <0.05). Gene *Rps3a3* is not shown. (**C)** Peripheral blood plasma levels of CXCL10 in wild-type and *Adar*$^{+/-}$ adult mice (n=12 age-matched pairs. Mean +/- s.d. **P=0.0019 paired t-test). (**D)** Western blot analysis of liver of wild-type and *Adar*$^{+/-}$ adult mice (Representative n=2 age-matched pairs). (**E)** *P. yoelii* parasitemia (% of *P. yoelii* iRBCs) following tail vein injection with 10^5 *P. yoelii* 17XNL:PyGFP infected RBCs monitored until resolution of infection (by 30 days) (n=6 age-matched female mice, mean +/- s.e.m *P=0.118, Two-way ANOVA with Geisser-Greenhouse correction).

a

Day 7 *P.yoelii*
***Adar+/-* _ wild type**
DE genes (Log2FC >1 and Adj.p-value <0.05)

Gene	Log2FC	Adj.p-value
Rps3a3	4.49	6.59E-136
Rps3a2	3.27	4.01E-43
Treml1	4.94	1.8E-27
Hist1h2al	8.58	3.97E-12
Htatip2	7.85	1.21E-10
Gm42743	6.29	3.66E-10

b



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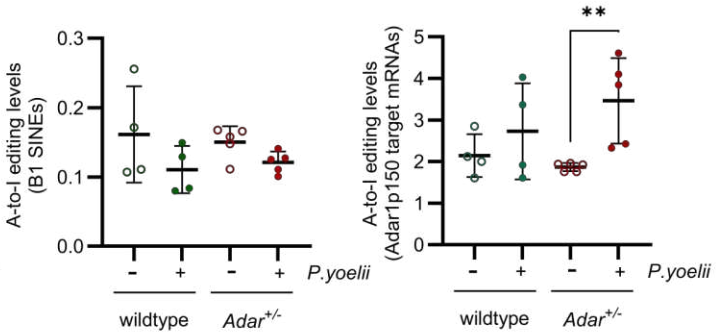

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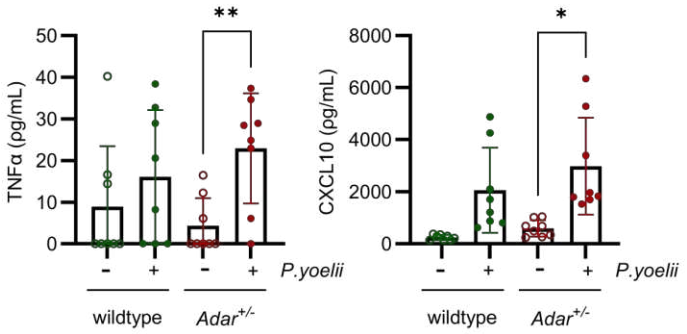

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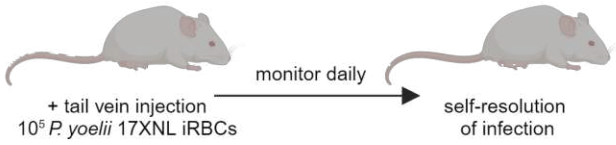

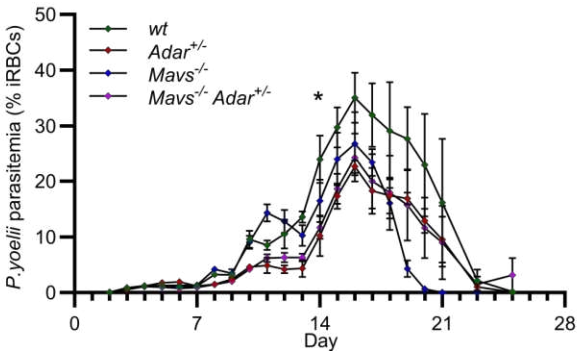

Fig. 3. *Adar*$^{+/-}$ mice are protected from *P. yoelii* parasitemia independently of the RLR pathway. (**A-B**) RNA sequencing analysis of whole blood of wild-type and *Adar*$^{+/-}$ adult mice on day 7 following infection with *P. yoelii* (n=3 age-matched pairs). (**A**) Table of differentially expressed (DE) genes comparing day 7 *P. yoelii*-infected *Adar*$^{+/-}$ mice with d7 *P. yoelii*-infected wild-type mice. (6 genes, Adjusted p-value <0.05, Log2 FoldChange>1). Genes that are also DE when comparing uninfected *Adar*$^{+/-}$ mice with uninfected wild-type mice are shown in grey, genes that are uniquely DE in this data set are shown in black. (**B**) Volcano plot of differentially expressed genes comparing d7 *P. yoelii Adar*$^{+/-}$ mice with d7 *P. yoelii* wild-type mice. Significantly DE genes shown in red (Adjusted p-value <0.05). (**C**) A-to-I RNA editing analysis of wild-type and *Adar*$^{+/-}$ adult mice from RNA-sequencing (n=4-5 age-matched pairs. Mean +/- s.d. (Left) B1 SINE A to G editing index. Wild-type n.s. P=0.3829, paired t-test (n=4); *Adar*$^{+/-}$ n.s. P=0.0549, paired t-test (n=5). (Right) ADAR1 p150 target mRNA A to G editing index. Wild-type n.s. P=0.5098, paired t-test (n=4); *Adar*$^{+/-}$ *P=0.0249, paired t-test (n=5)). (**D**) Cytokine levels of wild-type and *Adar*$^{+/-}$ mice following *P. yoelii* infection (d7). (n=8 age-matched pairs. Mean +/- s.d. CXCL10 wild-type n.s. P=0.0580, *Adar*$^{+/-}$*P=0.0200; TNFα wild-type n.s. P=0.3317, *Adar*$^{+/-}$ *P=0.0485. One-way ANOVA with Tukey's multiple comparison test). (**E**) *P. yoelii* parasitemia (% of *P. yoelii* iRBCs) following tail vein injection with 10^5 *P. yoelii* 17XNL:PyGFP iRBCs monitored until resolution of infection (by 30 days) (n=3-5 age-matched female mice, mean +/- s.e.m. Day14: wt vs *Adar*$^{+/-}$ *P=0.0277. wt vs *Mavs*$^{s-/-}$ *Adar*$^{+/-}$ *P=0.0280. Two-way ANOVA with Tukey's multiple comparison test).

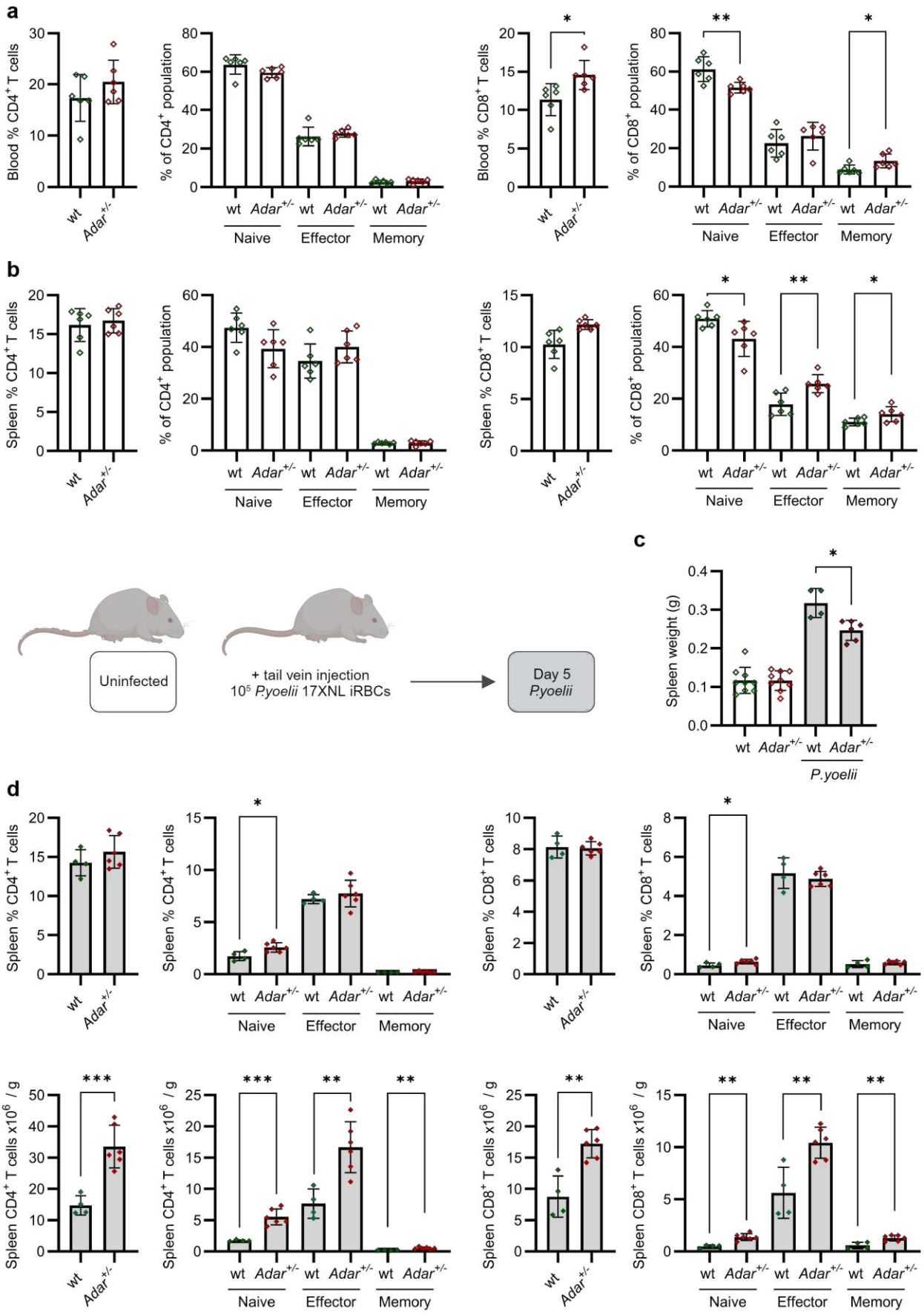

Fig. 4. *Adar*$^{+/-}$ mice display T cell phenotypes associated with protection from malaria. (**A**) Percentage (%) of T cells in circulating blood of wild-type and *Adar*$^{+/-}$ mice (n=6 age-matched pairs. Mean +/- s.d. Unpaired t-test.*P<0.05, **P<0.005). (**B**) Percentage (%) of T cells in spleen of wild-type and *Adar*$^{+/-}$ mice (n=6 age-matched pairs. Mean +/- s.d. Unpaired t-test). (**C**) Spleen weights of uninfected wild-type (n=9) and *Adar*$^{+/-}$ (n=9) mice (n.s), and *P.yoelii* infected (day 5) wild-type (n=4) and *Adar*$^{+/-}$ (n=6) mice (*P=0.0361, One-way ANOVA with Tukey's multiple comparison test). (**D**) Percentage (%) (upper panel) and total number of T cells per g spleen weight (lower panel) of *P.yoelii* infected (day 5) wild-type (n=4) and *Adar*$^{+/-}$ (n=6) mice (Mean +/- s.d. Unpaired t-test. *P<0.05, **P<0.005, ***P<0.0005).