

1 **Supplemental Information**

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3 **Gcn5 and PCAF regulate *PPAR $\gamma$*  and *Prdm16* expression to facilitate brown**  
4 **adipogenesis**

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10 **Figure S1. Gcn5 and PCAF function redundantly to regulate adipogenesis**

11 (A – C) Gcn5 and PCAF function redundantly during white adipocyte differentiation.

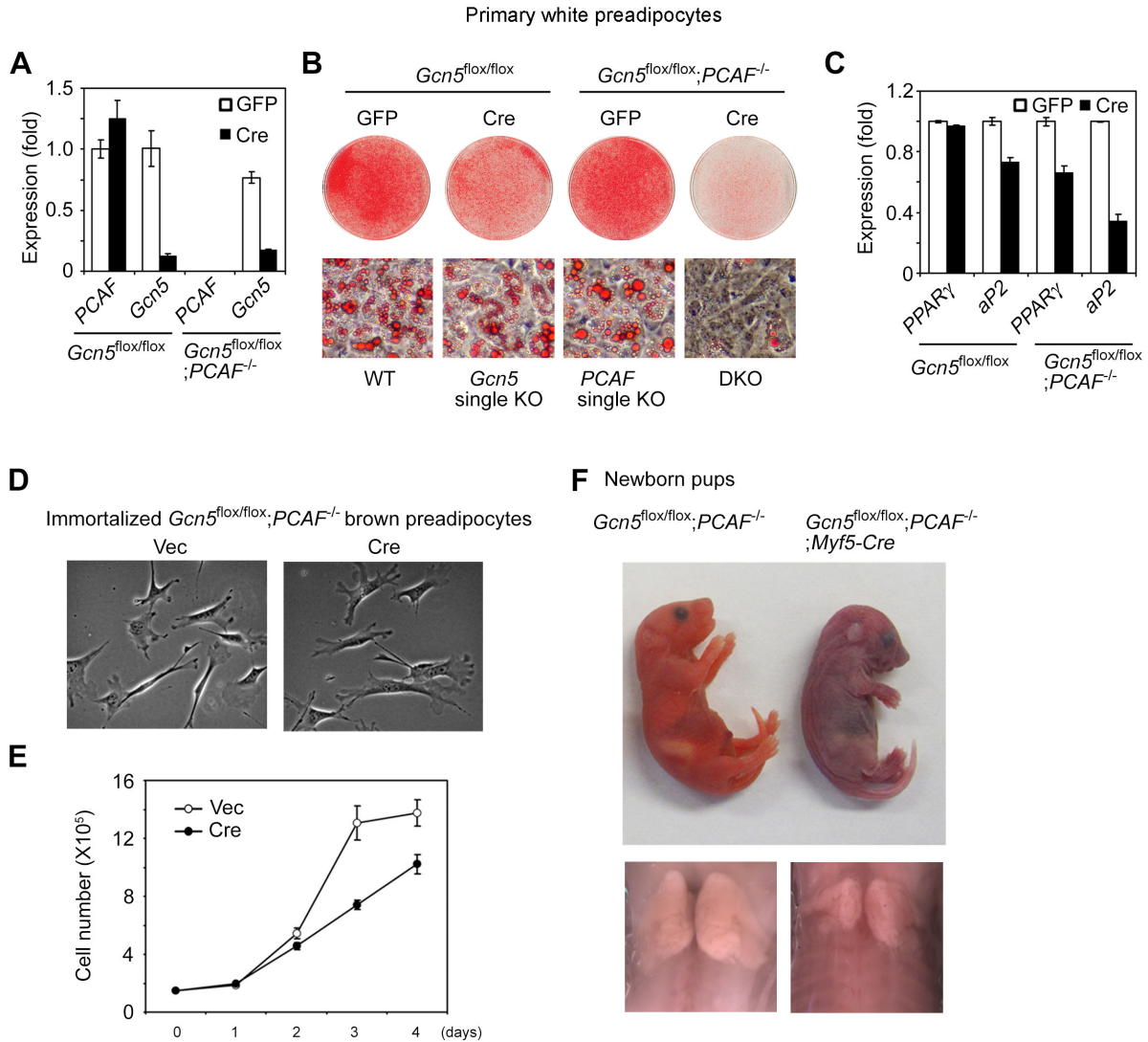
12 Subcutaneous white preadipocytes were isolated from *Gcn5*<sup>flox/flox</sup> and *Gcn5*<sup>flox/flox</sup>;*PCAF*<sup>-/-</sup>  
13 mice, and infected with adenoviral Cre to delete *Gcn5* to generate *Gcn5* single KO or  
14 *Gcn5/PCAF* DKO white preadipocytes. KO of *Gcn5* and *PCAF* was confirmed by qRT-  
15 PCR (A). The cells were induced to undergo adipogenesis for 6 days, followed by Oil  
16 Red O staining (B) and qRT-PCR analysis of gene expression (C).

17 (D – E) *Gcn5/PCAF* double deletion in brown preadipocytes has no effect on cell  
18 morphology, but retards cell growth. Retroviral Vec- or Cre-infected immortalized  
19 *Gcn5*<sup>flox/flox</sup>;*PCAF*<sup>-/-</sup> brown preadipocytes were analyzed for cell morphology under  
20 microscope (D). Cell growth curves are shown in (E).

21 (F) Gcn5 and PCAF are required for BAT development *in vivo*. *Gcn5*<sup>flox/flox</sup>;*PCAF*<sup>-/-</sup>  
22 males were crossed with *Gcn5*<sup>flox/+</sup>;*PCAF*<sup>-/-</sup>;*Myf5-Cre* females to generate

23 *Gcn5<sup>flox/flox</sup>;PCAF<sup>-/-</sup>;Myf5-Cre* pups. The *Gcn5<sup>flox/flox</sup>;PCAF<sup>-/-</sup>;Myf5-Cre* pups died  
 24 postnatally and exhibited smaller BAT.

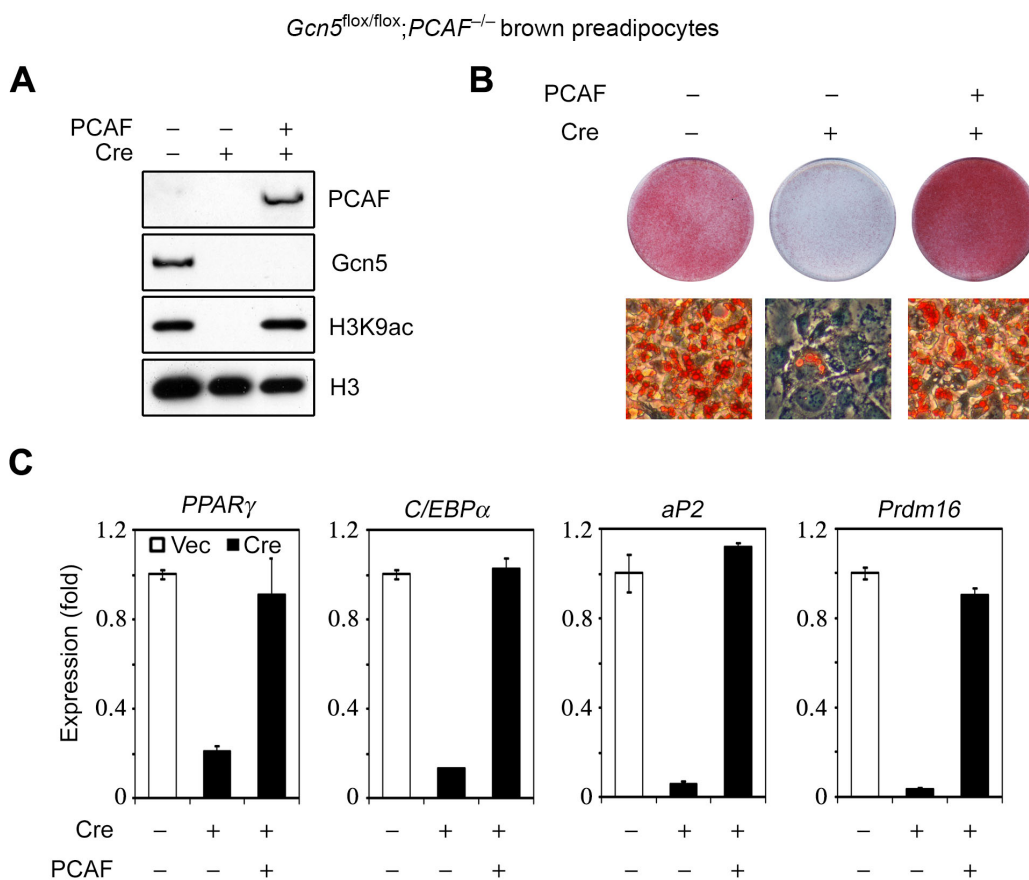
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**Figure S1. Gcn5 and PCAF function redundantly to regulate adipogenesis**

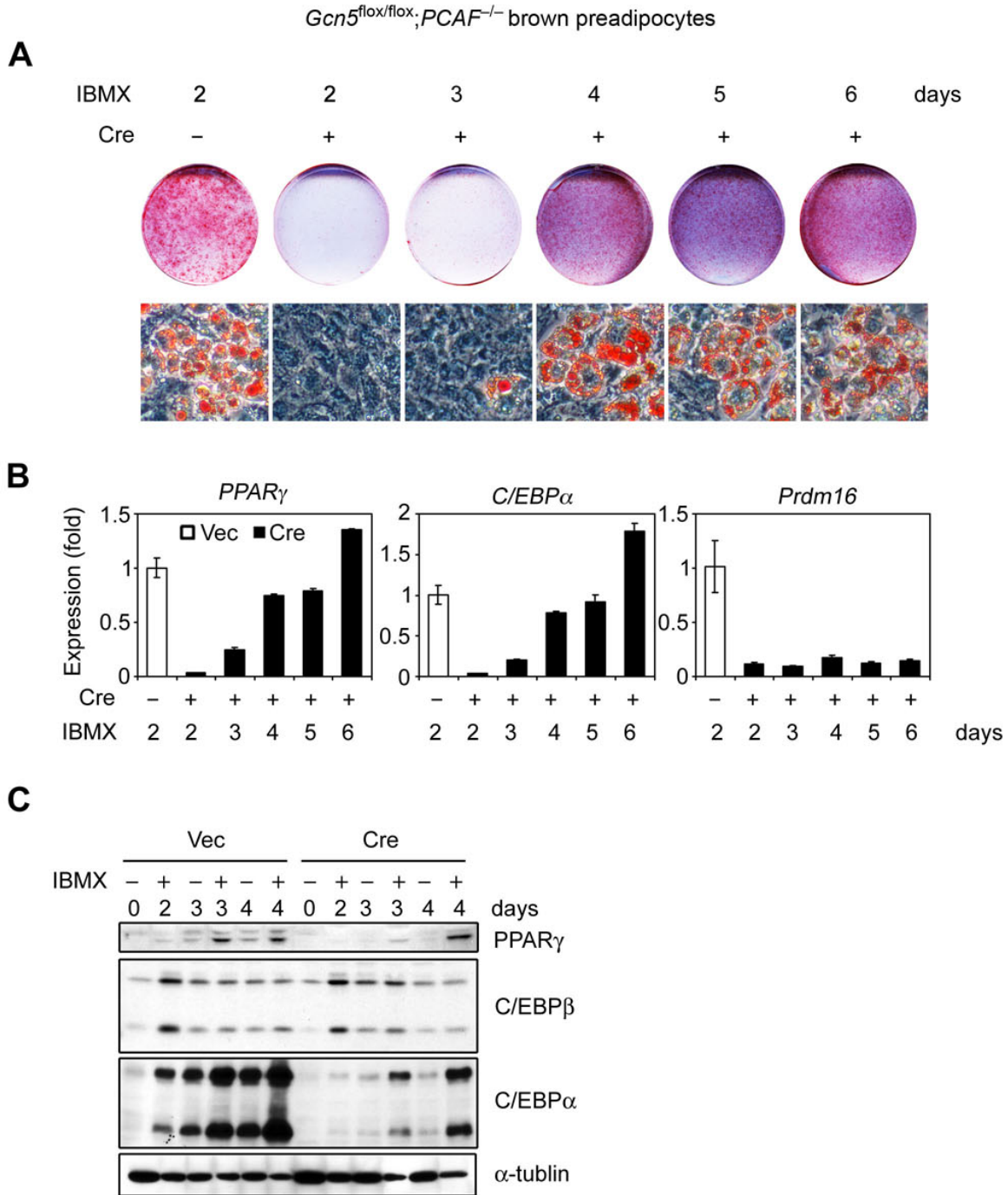
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27 **Figure S2. PCAF rescues loss of the global H3K9ac and defective adipogenesis in**  
 28 ***Gcn5/PCAF* DKO cells**  
 29 *Gcn5*<sup>flox/flox</sup>;*PCAF*<sup>-/-</sup> brown preadipocytes were sequentially infected with retroviruses  
 30 expressing PCAF and Cre. (A) PCAF, Gcn5, histone H3 and H3K9ac in subconfluent  
 31 cells were analyzed by immunoblotting. The cells were induced to undergo  
 32 adipogenesis for 6 days, followed by Oil Red O staining (B) and qRT-PCR analysis of  
 33 gene expression (C).  
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**Figure S2. PCAF rescues loss of the global H3K9ac and defective adipogenesis in *Gcn5/PCAF* DKO cells**

35 **Figure S3. Prolonged IBMX treatment rescues defective adipogenesis in**  
36 ***Gcn5/PCAF* DKO cells**  
37 Retroviral Vec- or Cre-infected *Gcn5*<sup>flox/flox</sup>;*PCAF*<sup>-/-</sup> brown preadipocytes were induced to  
38 undergo adipogenesis. After removal of the adipogenic cocktail at day 2, the cells were  
39 treated with 0.5 mM IBMX until the indicated day. The cells were collected at day 6 and  
40 subjected to Oil Red O staining (A), and qRT-PCR analysis of gene expression (B).  
41 (C) At day 3 and 4, the cells were treated with or without IBMX, and the cells were  
42 collected and subject to immunoblotting using antibodies indicated on the right.  
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**Figure S3. Prolonged IBMX treatment rescues defective adipogenesis in *Gcn5/PCAF* DKO cells**