

1 **Supplementary information**

2 **Orobol, an Enzyme-Convertible Product of Genistein, exerts Anti-Obesity Effects by Targeting Casein**  
3 **Kinase 1 Epsilon**

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7 **Supplement Figure 1. Effects of Orobol blocked the G1 phase during mitotic clonal expansion.** Cells  
8 were harvested at the indicated time after the initiation of differentiation with or without Orobol treatment and  
9 stained with PI solution for flow cytometer cell cycle analysis. The histogram results indicate cell cycle  
10 distribution in G1, S or G2/M phases. Data were represented as mean  $\pm$  SEM values of at least three  
11 independent experiments. To compare the differences among the groups, one-way ANOVA was used with  
12 Duncan's multiple range tests. Different letters indicated statistically significant differences at  $p < 0.05$ .

13  
14 **Supplement Figure 2. Effects of CK1 $\epsilon$  inhibitor on MDI-induced adipogenesis.** (A) After cell  
15 differentiation, 3T3-L1 adipocytes were stained with Oil red O and images were captured. (B) Quantification  
16 of intracellular lipid content. (C) Orobol does not exhibit cytotoxicity in 3T3-L1 preadipocytes up to 40  $\mu$ M.  
17 Data are represented as means  $\pm$  SEM from at least three independent experiments. The sharps (##) indicate  
18 a significant difference between the control group and the group treated with the MDI cocktail alone ( $p <$   
19  $0.01$ ). Means with different letters (a–d) within a graph are significantly different between groups treated with  
20 MDI alone and those treated with MDI plus PF5006739

21  
22 **Supplement Figure 3. Effects of orobol on MDI-induced signaling in 3T3-L1 preadipocytes.**

23 Western blot analysis for Akt, mTor and p70<sup>S6K</sup> in MDI-induced signaling in 3T3-L1 preadipocytes. The data  
24 are representative of three independent experiments that gave similar results.

25 **Supplement Figure 4.** Effects of orobol on the liver weight in HFD-fed mice. Orobol treatment for 23  
26 weeks decreased the weight of liver tissue in HFD-fed mice. Values are expressed as means  $\pm$  SEM. The  
27 sharp (#) indicates a significant difference between the control group and the group treated with HFD ( $p <$   
28  $0.05$ ). The means marked with '\*' indicate significantly different compared to the group treated with HFD ( $p$   
29  $< 0.05$ )  
30

1 **Supplement Fig.5.** These are uncropped blot images corresponding to data show in Figure 3.

2

3 **Supplement Fig.6.** These are uncropped blot images corresponding to data show in Figure 4A.

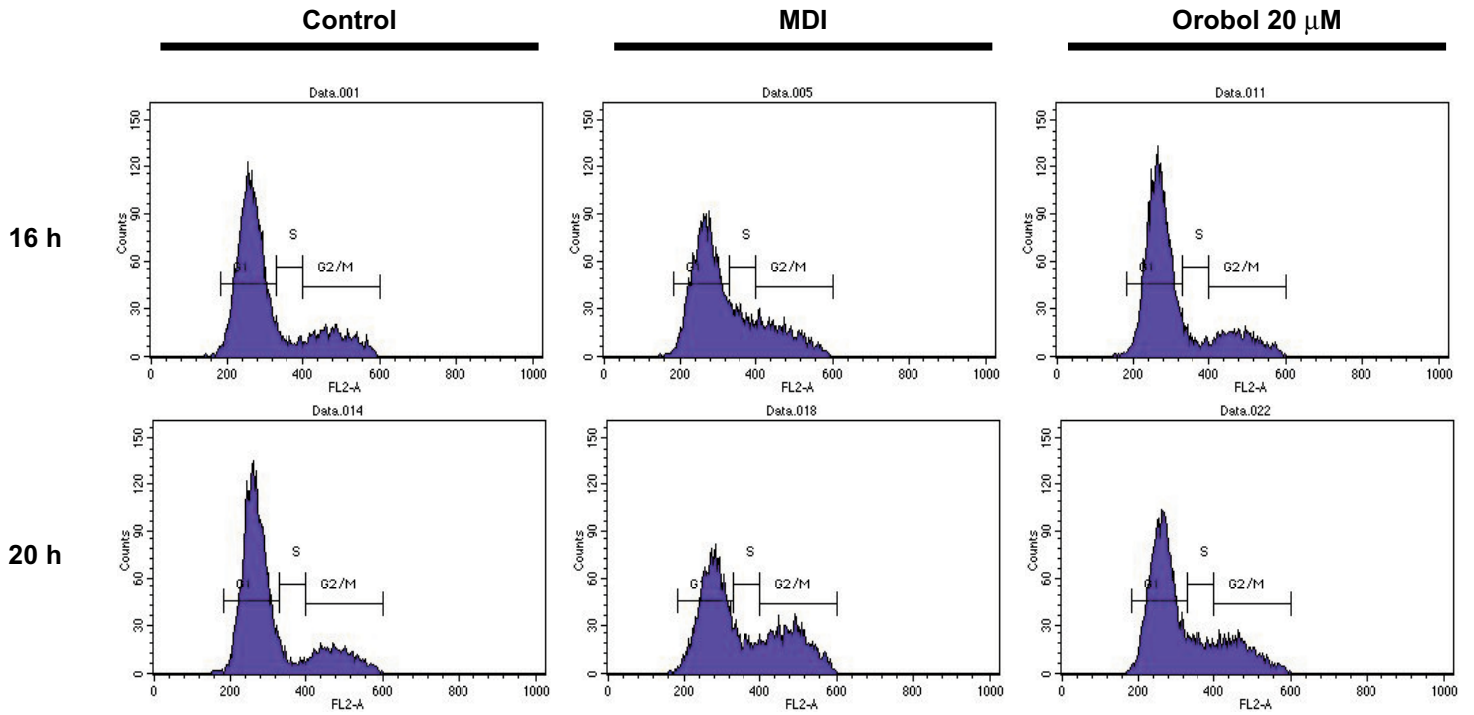
4

5 **Supplement Fig.7.** These are uncropped blot images corresponding to data show in Figure 4B.

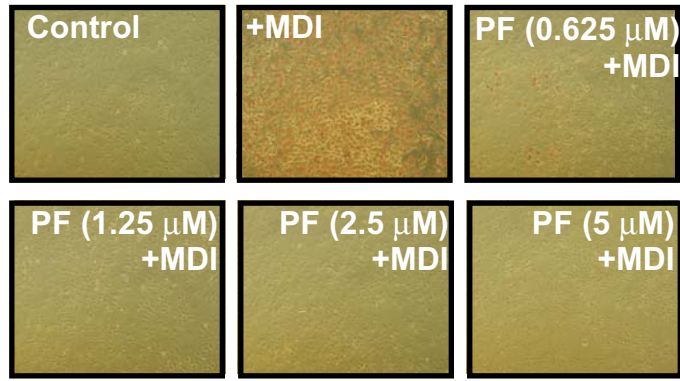
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7 **Supplement Table 1.** Inhibition of kinase activity by 20  $\mu$ M orobol

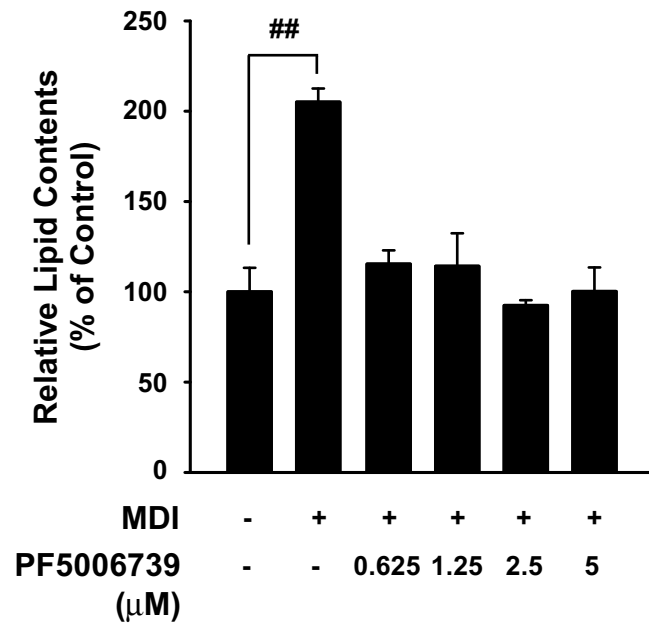
# Supplement Figure 1



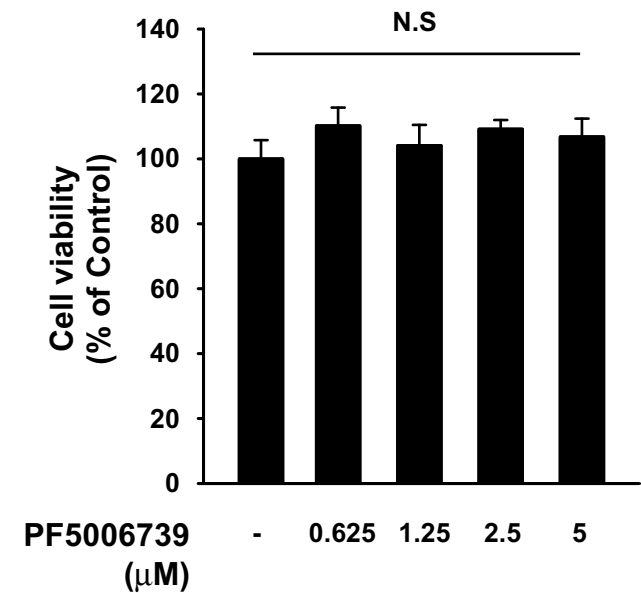
## A



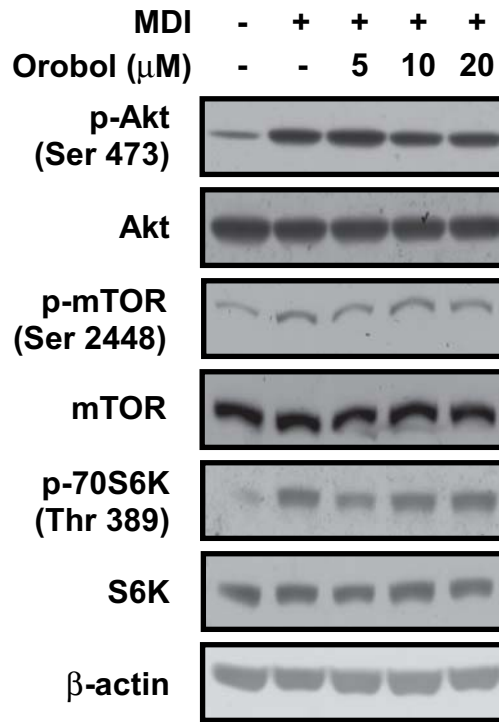
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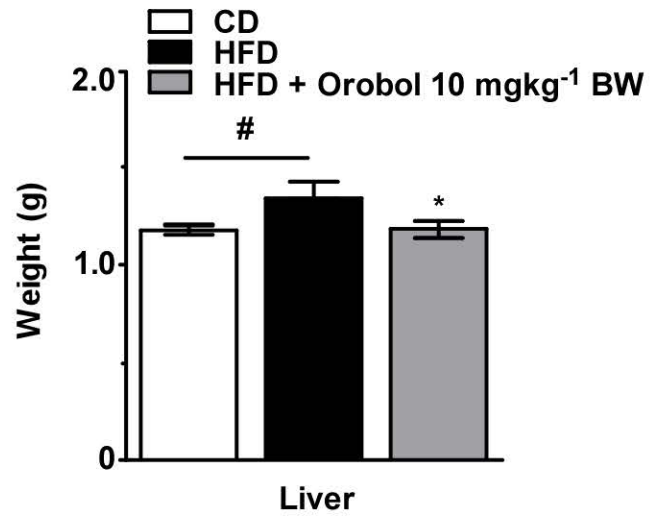
## C



# Supplement Figure 3



## A



# Supplement Figure 5

Figure 3B



Figure 3B

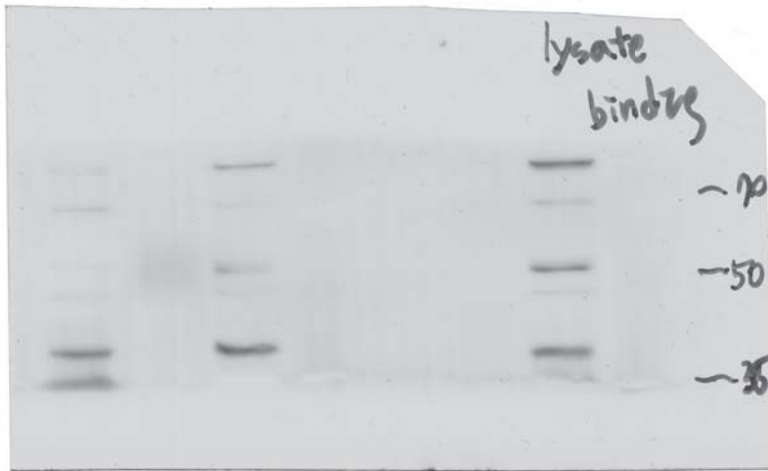
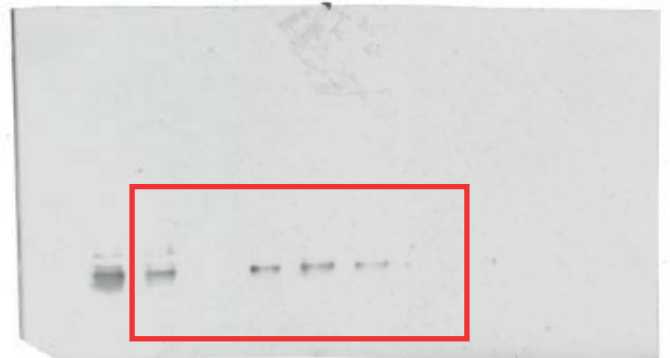
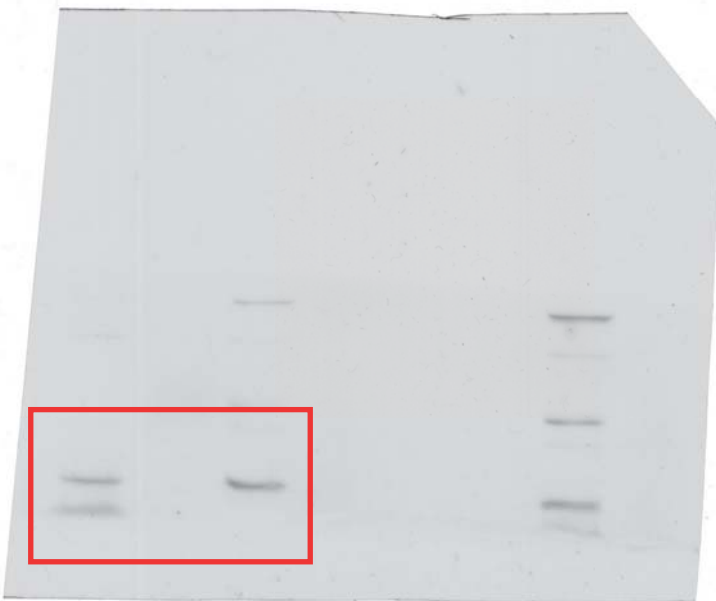


Figure 3B

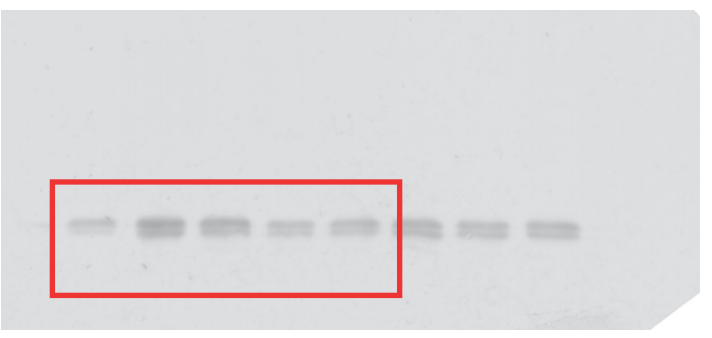




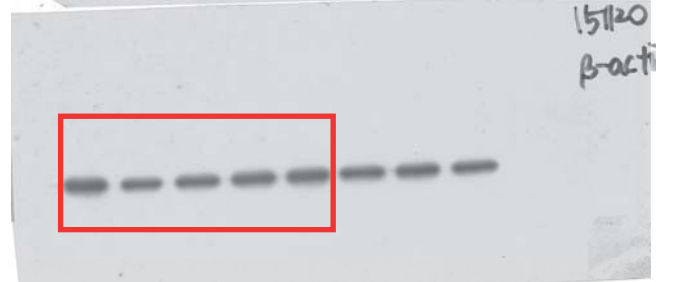
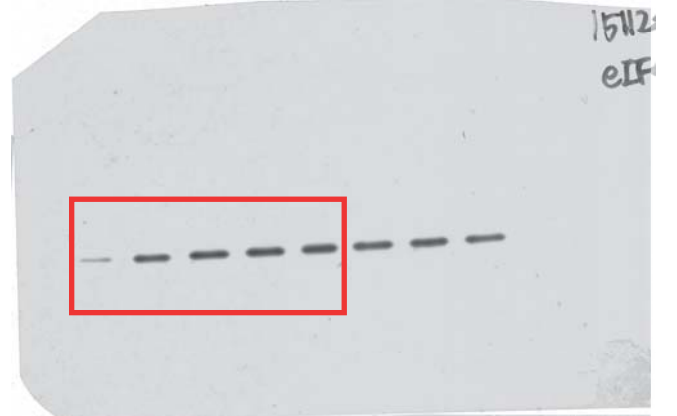
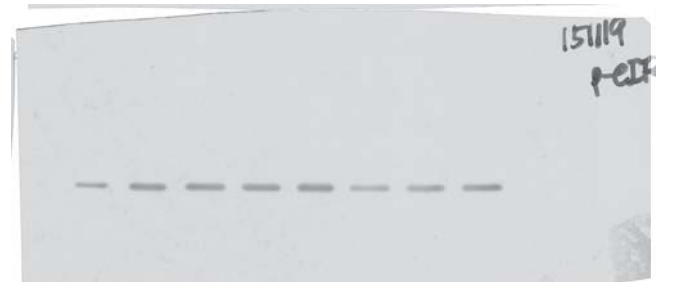
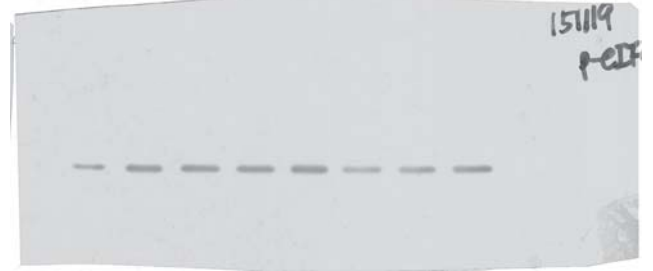
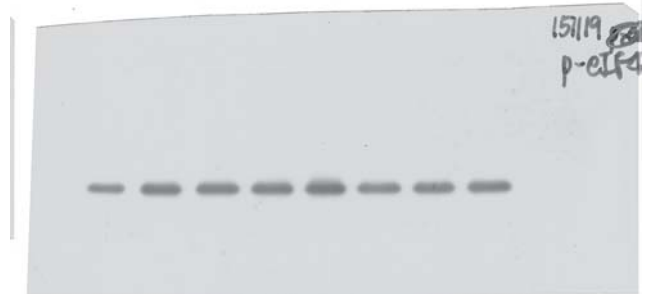
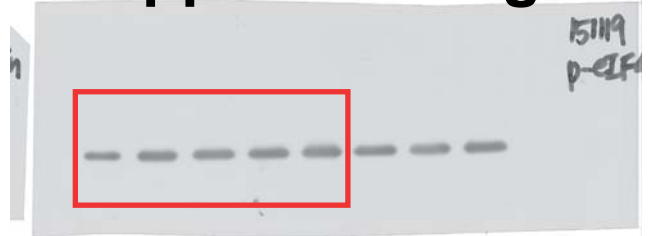
# Supplement Figure 6



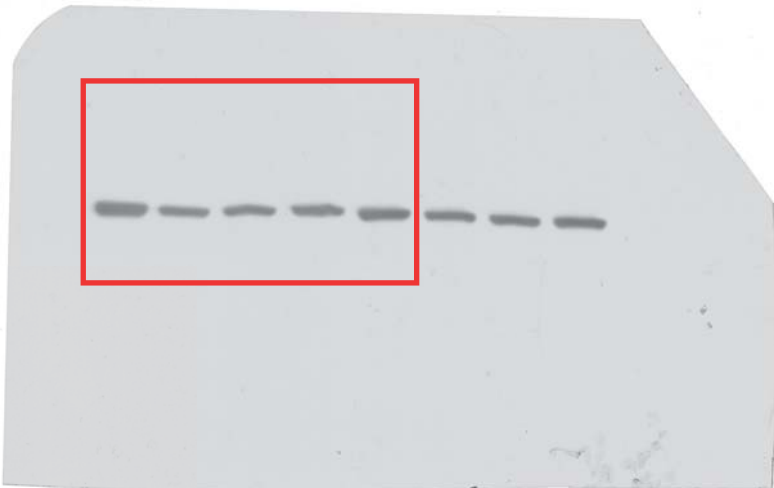
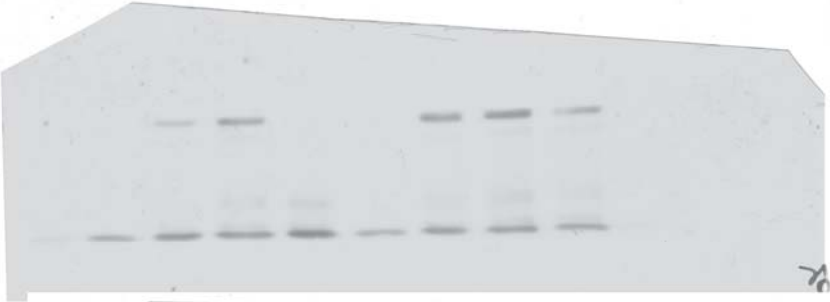
- 1. Control
- 2. MDI
- 3. Orobol 5 +MDI
- 4. Orobol 10 +MDI
- 5. Orobol 20 +MDI



- 1. Control
- 2. MDI
- 3. Orobol 5 +MDI
- 4. Orobol 10 +MDI
- 5. Orobol 20 +MDI



# Supplement Figure 7



# Supplement Figure 8

