Mice lacking PLAP-1/aspersin counteracts high fat diet-induced metabolic disorder and alveolar bone loss by controlling adipose tissue expansion

Hiromi Sakashita¹, Satoru Yamada¹,²*, Masaki Kinoshita¹, Tetsuhiro Kajikawa¹,⁸, Tomoaki Iwayama¹, and Shinya Murakami¹*

¹ Department of Periodontology, Osaka University Graduate School of Dentistry, Suita, Osaka, Japan
² Department of Periodontology and Endodontology, Tohoku University Graduate School of Dentistry, Sendai, Miyagi, Japan
⁸ Current Address: Department of Basic and Translational Sciences, Laboratory of Innate Immunity and Inflammation, Penn Dental Medicine, University of Pennsylvania, Philadelphia, Pennsylvania, United States of America

* Corresponding author

E-mail: satoruy@tohoku.ac.jp (SY) and ipshinya@dent.osaka-u.ac.jp (SM)
Fig S1
Fig S1. HFD-induced obesity and metabolic abnormalities.

(A) 5-week-old male C57BL/6J mice were fed with high fat diet (HFD) or normal chow diet (NC) and weighted weekly (n = 7 in each group). (B) Glucose tolerance test (GTT) and insulin tolerance test (ITT) were performed in mice after NC or HFD feeding (n = 4 in each group). (C) Serum levels of total cholesterol (T-CHO), triglyceride (TG), LDL-cholesterol (LDL-C), HDL-cholesterol (HDL-C), non-esterified fatty acid (NEFA), glucose (GLU) were measured during NC or HFD feeding (n = 3 in each group). *: $p < 0.05$, **: $p < 0.01$, ****: $p < 0.0001$. 
Fig S2

A

B

Weeks after feeding

NC

HFD

a

b

c

a+b+c (μm)

0 4 8 16

NC

HFD

---
Fig S2. HFD-induced alveolar bone resorption.

(A) Alveolar bone resorption in mice fed with NC or HFD was evaluated by μCT. (B) Distance between alveolar bone crest and cement-enamel junction was measured at distal root of first molar (a), mesial (b) and distal (c) root of second molar (n = 14 in each group). Results show the mean ± SD. *: p < 0.05
Fig S3

A

B

GTT

Blood glucose (mg/dL)

Time (min)

WT

Plap-1 KO

AUCGTT

WT

Plap-1 KO

C

ITT

Blood glucose (mg/dL)

Time (min)

WT

Plap-1 KO

AUCITT

WT

Plap-1 KO
Fig S3. Body weight change and glucose homeostasis in Plap-1 KO mice with NC feeding.

(A) Body weight changes in WT and Plap-1 KO mice during NC feeding. 5-week-old male WT and Plap-1 KO mice were fed with NC and weighted weekly. WT (n = 11), Plap-1 KO (n = 8). Results show the mean ± SD. (B) GTT and ITT were performed in 5-week-old male WT and Plap-1 KO mice (n = 9 in each group). (C) GTT and ITT were performed in WT and Plap-1 KO mice after 16 weeks NC feeding. For GTT, WT (n = 8), Plap-1 KO (n = 7) and for ITT, WT (n = 8), Plap-1 KO (n = 8). Results show the mean ± SD. *: p < 0.05, **: p < 0.01, ***: p < 0.001, ****: p < 0.0001.
Fig S3. Immunoblots performed in this study

(A) anti-FLAG blot (left) and Ponceau S staining (right).
| Gene of interest | Forward primers sequences | Reverse primers sequences |
|------------------|---------------------------|---------------------------|
| Aspn             | atgtagcagataacgtagctgcaga | ttggttgggaacccgatgtcaga  |
| Adipoq           | ttctgtctgtcattgtagctgagga | gcgcgactgggactgggattta   |
| Lep              | gaacgcgtctctgcatgcacgca   | ctggctctgtagcctgttgg     |
| Adgre1           | gcgtggggctccagctgctgtgtg | gaaggtcagcaacctcgtgtctgcc |
| Tnf              | aagccttgagccccacgtcagta  | gcgcacactgtggtggtgcttttg |
| Il6              | ccactcacaagctggagcattta  | gcacgtgcatcagctgctctctaca|
| Ccl3             | catgacactctgcaacacagtttc | gcagcaagcctgtgggtttgcttc |
| Arg1             | agctctgggaatctgcatgg    | atgtacacgatgtctttggcagata|
| Chil3            | atgggcctcacaactggactg    | agtcaatggctctgtcccttg    |
| Clec7a           | ggtaacctgactcaaatcctcacaac | caccagatgatagtggctgttc |
| Fabp4            | cgggaaccttggaagctggcttc | gaattccacgccccacttgga   |
| Pparg            | ggagcctagttgtgtgtgctttg | tgcagcagttgctctggctg    |
| Cebpa            | cggagccagctggcagctggcagc | gcacactgcatctggaactaag |
| Col1a            | cagggtattgtcggagcaactgg | gcacctgttggcagacgtca    |
| Col3a1           | caggccaggtggcagctgtaaga | cccttgccttgctgttggata-  |
| Col6a1           | gagcttcctgagcagccccctc  | gagctggctcagggagcttgag  |
| Dcn              | ctgggctggcagcataagta    | cggacaggggtggccgtaaag   |
| Bgn              | cggagctgagatggacctgta   | tccgaagccccataaggacagaag|
| Hprt             | tggcggaagctggctagcagact | aggccgagctggccagagcagaag|