

SUPPLEMENTAL INFORMATION:

**Low ketolytic enzyme levels in tumors predict ketogenic diet responses
in cancer cell lines in vitro and in vivo**

Jie Zhang^{1, 5#}, Ping-Ping Jia^{2#}, Qing-Le Liu^{3#}, Ming-Hua Cong⁴, Yun Gao⁵, Han-Ping Shi^{2*},
Wei-Nan Yu^{1*}, Ming-Yong Miao^{5*}

¹Department of Endocrinology, Huai'an Hospital Affiliated to Xuzhou Medical University, and Huai'an Second People's Hospital, Huai'an 223002, Jiangsu, China. ²Department of Gastrointestinal Surgery / Clinical Nutrition, Beijing Shijitan Hospital, Capital Medical University, Beijing 100038, China. ³Department of Hyperbaric Oxygen Therapy, The First Affiliated Hospital of The Second Military Medical University, Changhai Hospital, Shanghai 200433, China. ⁴Department of Comprehensive Therapy, Cancer Hospital of Chinese Academy of Medical Sciences, Beijing 100021. ⁵Department of Biochemistry and Molecular Biology, The College of Basic Medical Sciences, The Second Military Medical University, Shanghai 200433, China.

Table S1. The exact composition of the diets

| | Standard diet (%) | Ketogenic diet (%) |
|--------------------------|--|---------------------------|
| Protein | 19.0 | 18.2 |
| Fat | 4.0 | 62.8 |
| Carbohydrate | 50.0 | 2.6 |
| Fiber | 5.0 | 4.8 |
| Ash content | ≤8.0 | 5.2 |
| Water content | <10.0 | <10.0 |
| others | vitamins ¹ and trace elements | |
| Calories per gram | 6.49kcal | 3.12kcal |

Note: ¹The exact composition of vitamins are shown in table S2.

Table S2. The exact composition of the vitamins

| | Value | Unit |
|------------------|--------------|-------------|
| Vitamin A | 13800 | 1U/kg |
| Vitamin B12 | 19 | mcg/kg |
| Ascorbic Acid | 0 | mg /kg |
| Vitamin D3 | 1900 | 1U/kg |
| Vitamin E | 220 | 1U/kg |
| Vitamin K3 | 2 | mg/kg |
| Biotin | 0.38 | mg /kg |
| Choline | 1790 | mg /kg |
| Folic acid | 3.8 | mg/kg |
| nicotinic acid | 57.1 | mg /kg |
| Pantothenic acid | 28 | mg /kg |
| Pyridoxine | 11 | mg /kg |
| Rinoflavin | 11.5 | mg /kg |
| Thiamine | 11.4 | mg/kg |

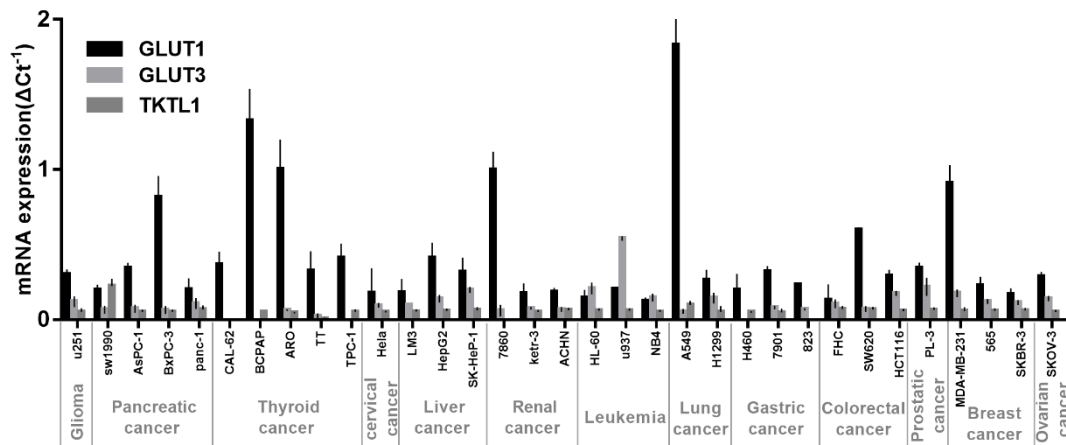


Fig. S1. The mRNA expression levels of relevant glycometabolic markers in 33 cancer cells by qRT-PCR.

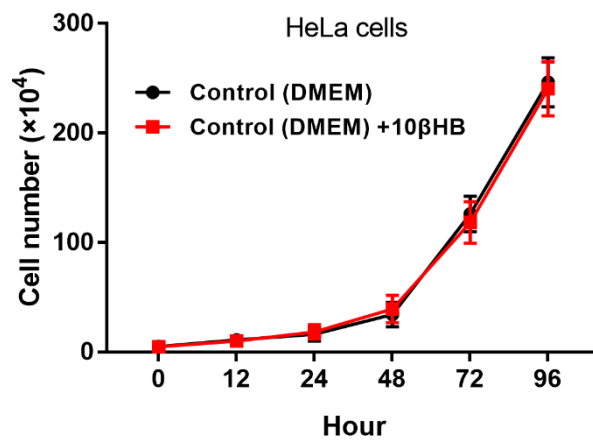


Fig. S2. Supplementation of control medium with 10 mMβHB does not affect proliferation of HeLa cells.

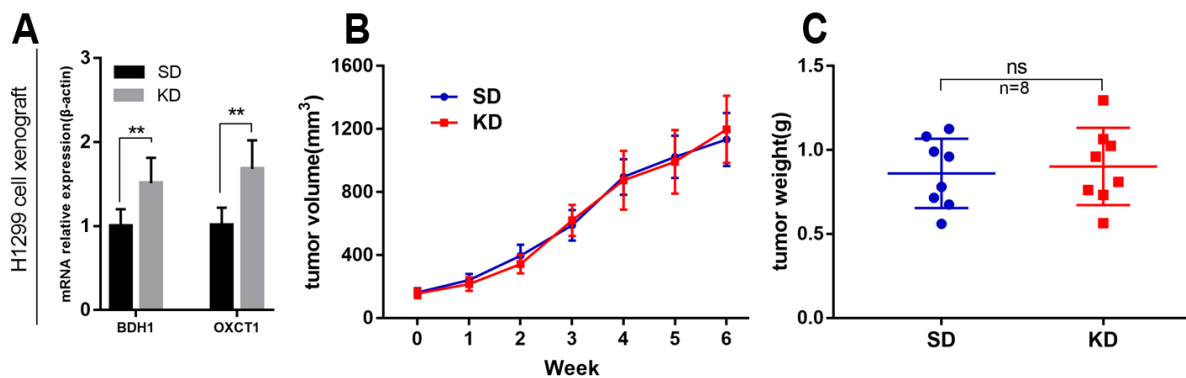


Fig. S3. KD could not inhibit tumor growth in H1299 cell xenografted nude mice. (A) The expression levels of BDH1 and OXCT1 mRNA in xenograft tumors by qRT-PCR. The relative quantitative expression was calculated by the $2^{-\Delta\Delta C_t}$ method. $**p < 0.01$ compared with the control. (B) Tumor growth of xenografted nude mice. (C) Tumor weight of xenografted nude mice.

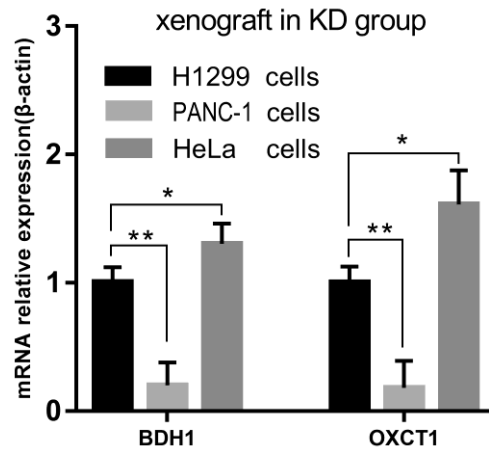


Fig. S4. The expression levels of BDH1 and OXCT1 in PANC-1, H1299 and HeLa cell xenografts in KD group. The BDH1 and OXCT1 expression levels in H1299 cell xenograft were lower than the ones in HeLa cell (* $P < 0.05$), but significantly higher than the ones in PANC-1 xenograft (** $P < 0.01$).